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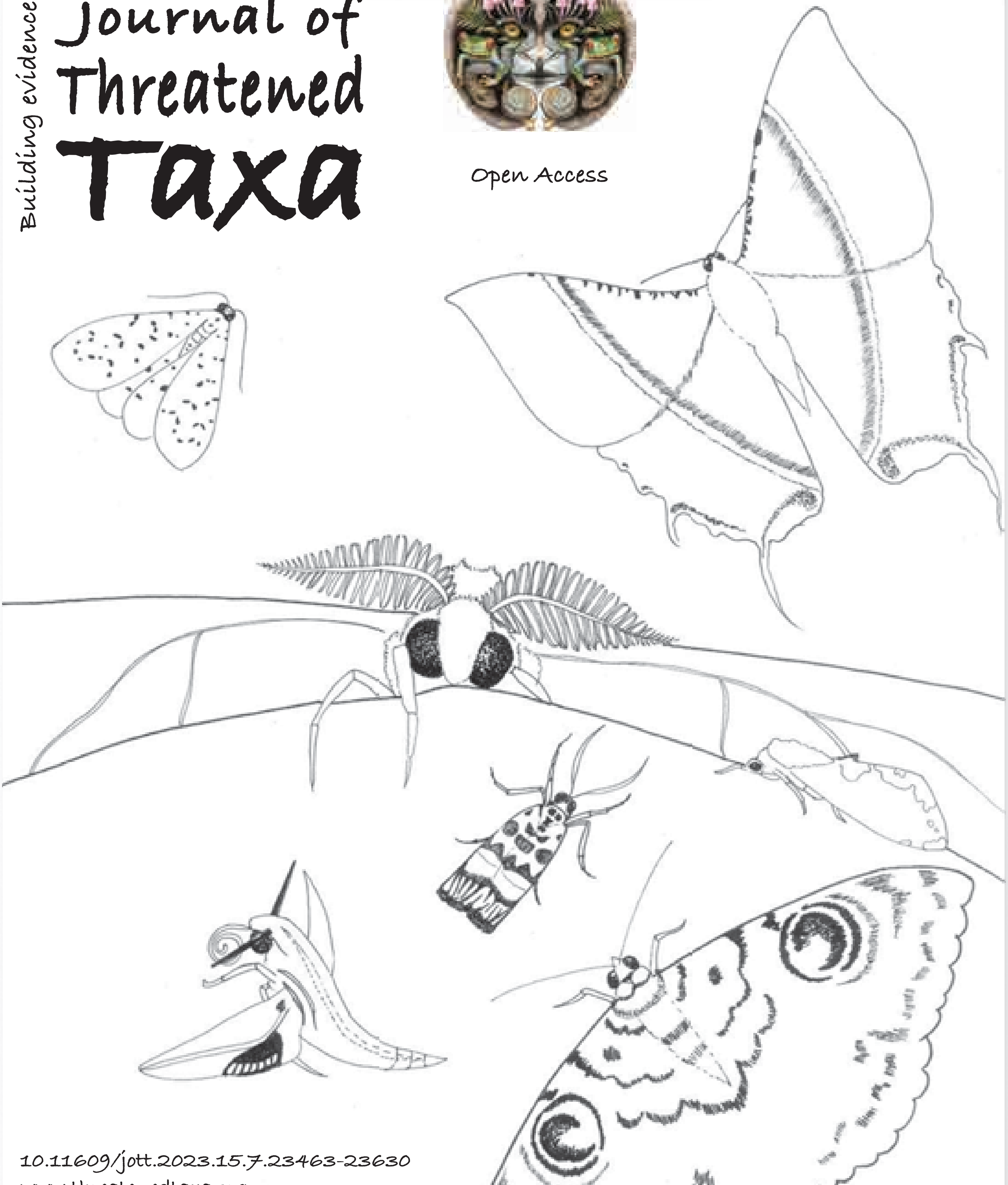

10.11609/jott.2023.15.7.23463-23630
www.threatenedtaxa.org

26 July 2023 (Online & Print)
15(7): 23463-23630
ISSN 0974-79t07 (Online)
ISSN 0974-7893 (Print)

ISSN 0974-7907 (Online); ISSN 0974-7893 (Print)

Publisher
Wildlife Information Liaison Development Society
www.wild.zooreach.org

Host
Zoo Outreach Organization
www.zooreach.org

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Cover: Celebrating the unsung heroes—moths, our nocturnal pollinators. © Priyanka Iyer.

Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23463–23471
ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)
https://doi.org/10.11609/jott.8291.15.7.23463-23471
#8291 | Received 01 December 2022 | Final received 17 June 2023 | Finally accepted 30 June 2023

ARTICLE

Predicting suitable habitat for the endangered Javan Gibbon in a submontane forest in Indonesia

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Abstract: Species distribution modeling is an essential tool for understanding the ecology of species and has many applications in conservation. Using maximum entropy (MaxEnt) modeling, we identify the key factors shaping the potential distribution of the endangered Javan Gibbons *Hylobates moloch* in one of the main remnant habitats, Gunung Halimun Salak National Park (GHSNP), Indonesia, using presence-only data collected between October and November 2015, and in April and May 2016. Maxent results showed that forest canopy density and annual temperature were the principal variables predicting the distribution of Javan Gibbons, with contribution scores of 53.9% and 35.6%, respectively. The predictive distribution map indicated that suitable habitat for Javan Gibbons is not uniformly distributed within GHSNP, i.e., suitable habitat is not located evenly throughout the region, with some areas more suitable than others. Highly suitable habitat comprises the largest proportion of habitat, with 42.1% of GHSNP classified as highly suitable habitat, whereas 24.7% was moderately suitable, and 33.2% of habitat was of low suitability for Javan Gibbons. Priority should be given to increasing habitat quality in degraded areas and law enforcement patrols to reduce degradation in peripheral regions of the park as part of the conservation management strategy.

Keywords: Conservation, forest canopy, *Hylobates moloch*, maximum entropy, West Java.

Abbreviations: AUC—Area under the curve | DEM—Digital elevation model | GHSNP—Gunung Halimun Salak National Park | IUCN—International Union for Conservation of Nature and Natural resources. | MaxEnt—Maximum entropy | MoEF—Ministry of Environment and Forestry | SDM—Species distribution models | PCA—Principal components analyses | ROC—Receiver operating characteristic | SRTM—Shuttle radar topography mission.

Editor: Mewa Singh, University of Mysore, Mysuru, India. **Date of publication:** 26 July 2023 (online & print)

Citation: Oktaviani, R., A. Borzée, A.N. Cahyana, S. Lappan, A. Mardiastuti & M.S. Giri (2023). Predicting suitable habitat for the endangered Javan Gibbon in a submontane forest in Indonesia. *Journal of Threatened Taxa* 15(7): 23463–23471. https://doi.org/10.11609/jott.8291.15.7.23463-23471

Funding: This work was funded by The Rufford Foundation, with grant number: 16654-1, and Idea Wild provided equipment for the field surveys.

Competing interests: The authors declare no competing interests.

Author details, Author contributions & Indonesian abstract: See end of this article

Acknowledgements: We thank the authorities of Gunung Halimun Salak National Park for granting us research permission (SIMAKSI No. 05/P/TNGHS/2015 and SIMAKSI No. 09/P/TNGHS/2016) and for their assistance during the field surveys. We thank the following individuals for their contributions to the fieldwork: A.M. Putra, Arya F., Djamaludin, E.M. Aaf Afnan, Fahri Budiman, Hery Sudarno, Kuntoro Bayu Aji, M. Faesal R.K., M. Choerudin, M. Sukri, Putra Caeruleus, and Yanuar I.D. We also thank Hariyo T. Wibisono, the editor, and the anonymous reviewers for their valuable comments and suggestions that improved the manuscript.

INTRODUCTION

Understanding the distribution of animals in space and ecological predictors of abundance are crucially important for designing effective conservation plans (Sarma et al. 2015). However, for most species, resources are not adequate to permit detailed surveys across every area of their potential distribution range. To address this problem, various modeling techniques have been developed to predict species distributions and identify suitable habitats by combining occurrence records with digital layers of environmental variables (Peterson 2001; Guisan & Thuiller 2005; Ortega-Huerta & Peterson 2008). Species distribution models (SDM) have been applied to various conservation problems. For instance, SDM have been used to prioritize areas for conservation (Araújo & Williams 2000), to predict geographical patterns of species occurrence (Peterson 2003), to discover unknown populations (Pearson et al. 2006), to improve the assessment of risk status (Solano & Feria 2006), and to predict species displacement patterns resulting from climate change (Borzée et al. 2019).

Several algorithms for modeling distributions use evidence of the presence or absence of a species in different locations. However, reliably determining that a species is absent is not often possible, limiting these algorithms' applicability. Alternatively, maximum entropy models (MaxEnt) aim to characterize species probability distributions using presence-only data, and can be applied even in situations with incomplete information from limited datasets (Pearson et al. 2006; Phillips et al. 2006; Guisan et al. 2007). MaxEnt can accurately predict habitat suitability based on relatively few variables (Liu et al. 2001; Dayton & Fitzgerald 2006) and these models can conform to the realized niche of species (Stone et al. 2013). This approach has been used to develop SDM in a wide range of primate species, including Asian Slow Lorises *Nycticebus* spp. (Thorn et al. 2009), Spider Monkey *Ateles geoffroyi* (Vidal-García & Serio-Silva 2011), Ecuadorian Capuchin *Cebus albifrons* (Campos & Jack 2013), Peruvian Night Monkey *Aotus miconax* (Shanee et al. 2015), Eastern Hoolock Gibbon *Hoolock leuconedys* (Sarma et al. 2015), Western Hoolock Gibbon *Hoolock hoolock* (Naher et al. 2021), Southern Yellow-Cheeked Gibbon *Nomascus gabriellae* (Nhung et al. 2021), and Bornean Agile Gibbon *Hylobates albibarbis* (Singh et al. 2018).

Javan Gibbons *H. moloch* are endemic to Java, Indonesia, and are generally restricted to the western and central parts of the island (Nijman 2004). Globally, Javan Gibbons are listed as 'Endangered' on the IUCN Red List (Nijman 2020). This species is sensitive to habitat alteration because of their dependence on closed-canopy forests for food (Kim et al. 2012), locomotion (Bertram 2004), and sleeping trees (Ario et al. 2018). Deforestation and forest degradation are primary threats as they disrupt the forest canopy and result in habitat fragmentation (Geissmann 2003; Smith et al. 2017).

It is estimated that up to 96% of the original Javan Gibbons habitat has been lost (Supriatna 2006; Nijman 2013; Malone et al. 2014), and most of the remaining habitat is located in protected areas such as Gunung Halimun Salak National Park (GHSNP). GHSNP is the largest remaining forest block in the region and represents the last stronghold for the species, likely harboring 25% and 50% of the global Javan Gibbon population (Nijman 2004). However, estimates of the total population within GHSNP vary dramatically, and populations within GHSNP may be effectively isolated from each other by enclaves of human activity within the park. The probability of persistence for these populations in the long term is likely to be affected by the total carrying capacity and the degree of isolation among subpopulations within GHSNP (Smith et al. 2017). Therefore, a better understanding of the total carrying capacity of GHSNP and the factors affecting habitat suitability is critical for effective conservation planning.

Two habitat suitability analyses for Javan Gibbons in GHSNP have been conducted using principal components analyses (PCA). Helianthi et al. (2007) estimated that 71.43% of the total area of GHSNP is highly suitable for Javan Gibbons, while Ikbal et al. (2008), in an analysis restricted to the Mount Salak region within GHSNP, estimated that only 13.20% of the habitat was highly suitable. Given changes in forest management and ongoing habitat alteration, habitat quality for Javan Gibbons in GHSNP may have changed in recent years; thus, a new approach and update are needed. We used MaxEnt modeling to identify environmental factors that contribute to the Javan Gibbon presence and to identify areas in GHSNP where habitat characteristics best align with the ecological niche of the species. The results of this study may help identify priority areas for conservation efforts and may lead to improved management practices within the park to ensure the continued survival of Javan Gibbons as one of the key species in GHSNP.

MATERIALS AND METHODS

Study Area

This study was conducted at GHSNP, Indonesia (6.739° S, 106.530° E), located within three administrative districts: Bogor and Sukabumi in West Java Province and Lebak in Banten Province. The Halimun area was established as a national park in 1992. To reduce forest loss, the Indonesian government increased the size of the protected area in 2003 by merging Halimun National Park and Salak Reservation Area, including the production forest. Currently, GHSNP covers an area of approximately 87,699 ha. Besides protecting water catchment areas for several big cities near the national park, it also protects essential habitat for endangered species such as Javan Gibbons, Javan Leopards *Panthera pardus melas*, and Javan Hawk-Eagles *Nisaetus bartelsii*. The park includes forests ranging from 500–2,200 m, a tropical climate with annual temperatures between 19° C and 31° C, and average precipitation of 4,000–6,000 mm. This national park experiences various pressures, including illegal gold mining, poaching, and forest encroachment for agricultural land & settlements, which cause fragmentation and degradation. Forest encroachment for agriculture is the biggest threat to GHSNP, driving fragmentation that may threaten the persistence of protected species in the area (Iwanda et al. 2019). Moreover, social conflicts related to land ownership, intensive land use, and ongoing timber exploitation by the rural community are significant problems in managing this national park (Rosleine et al. 2014).

Method

Field Survey

We conducted field surveys to determine the occurrence of Javan Gibbons at 10 locations across the GHSNP (Figure 1). We selected survey areas by combining historical information from Ikbal et al. (2008) and information obtained during a meeting in October 2015 with two GHSNP officers: Mr. Wardi Septiana from Conservation Area Affairs and Mr. Momo Suparmo from Biodiversity Conservation Affairs. In total, we obtained 73 occurrence records of Javan Gibbons across 10 survey sites representing ten resorts (the smallest administrative unit of the national park); 80.8% of occurrence records were based on direct observation, and 19.2% were based on indirect observation.

Field surveys were conducted in both rainy and dry seasons. The survey for the rainy season was undertaken between October and November 2015, while the dry season survey took place between April and May 2016 along the transect lines. To minimize negative impacts on the survey area, the survey team (2–3 people for each site, including at least one of the authors) walked along existing trails in the forest for 1–2 km depending on the difficulty of the terrain. Surveys were conducted for four hours in the morning (0700–1100 h) and three hours in the afternoon (1400–1700 h) each day of a four-day survey. This schedule was followed during both seasons except on heavy rainy days when we stopped the observation and repeated it the next day. The survey times were chosen based on the activity patterns of the species. During the walks, we recorded the time and location for all direct (visual) and indirect (auditory) encounters using a GPS Garmin 64s (Kansas, United States), by estimating the distance from the observers the individuals sighted by using Bushnell Digital Laser Rangefinder 850 (Utah, United States), and sighting angle between the transect line and the observers to species line.

Data Analysis

We included seven environmental variables in our models that were also used in previous modeling for the same species (Helianthi et al. 2007; Suheri et al. 2014; Widyastuti et al. 2020), and as they were found to be likely to influence habitat use by Javan Gibbons (Table 1).

We used MaxEnt v3.3.3 (Phillips et al. 2006) to produce a map of suitable habitats for Javan Gibbons in GHSNP. Of the 73 occurrence data points, 75% of points were used as a training sample and 25% of points as references for model validation. Environmental variables that predicted >10% of the variance in gibbon presence in the models were identified as important, following Norris et al. (2011).

We classified habitat with values < 0.25 as having low suitability, values between 0.25–0.75 as having moderate suitability, and values >0.75 as having high habitat suitability for Javan Gibbons. In most cases, values greater than 0.5 indicate suitable habitat (Yang et al. 2013). The default value of 1 has been identified as the most suitable to prevent overfitting (Merow et al. 2013).

Model accuracy should be tested in a modeling approach to evaluate model performance. We used a receiver operating characteristic (ROC) value closer to 1 to assess the model. This method does not require arbitrary threshold selection and has been widely used. The ROC generates a single measure of model performance called area under the curve (AUC) with AUC values >0.9 indicating high accuracy of the model (Elith et al. 2006; Phillips et al. 2006).

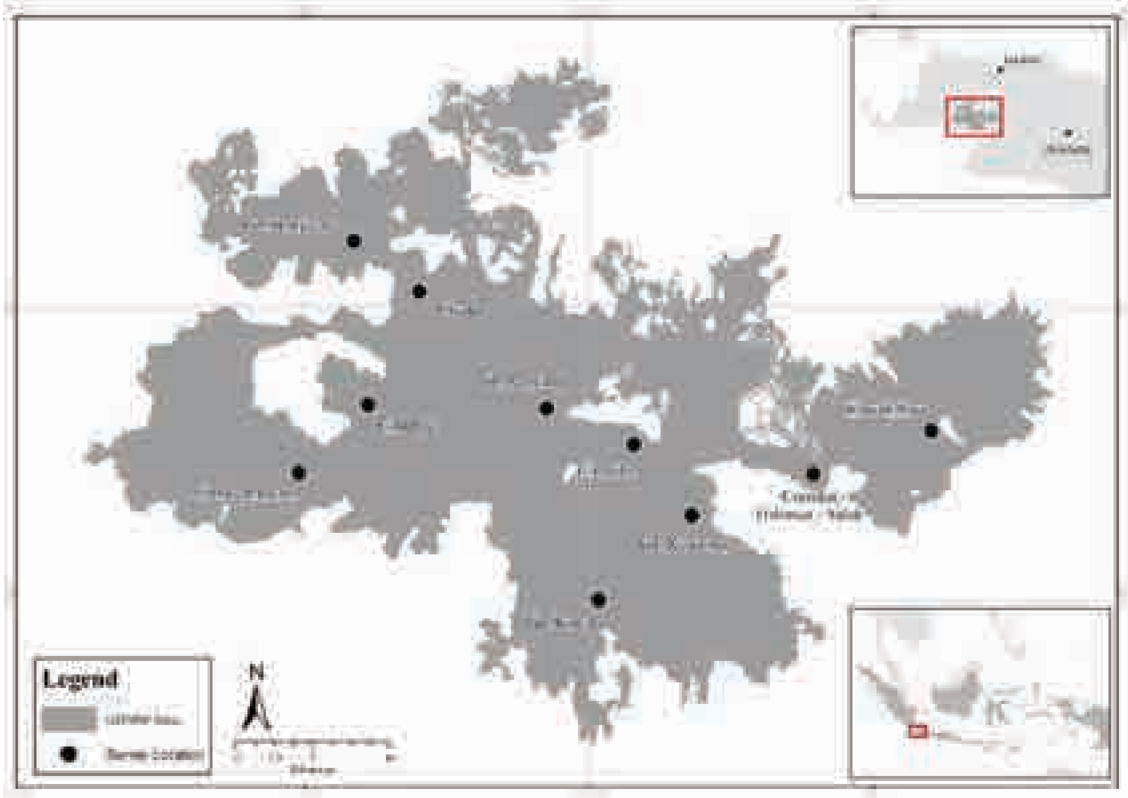

Figure 1. Locations in GHSNP, Indonesia, surveyed for the presence of Javan Gibbons in October–November 2015 and April–May 2016.

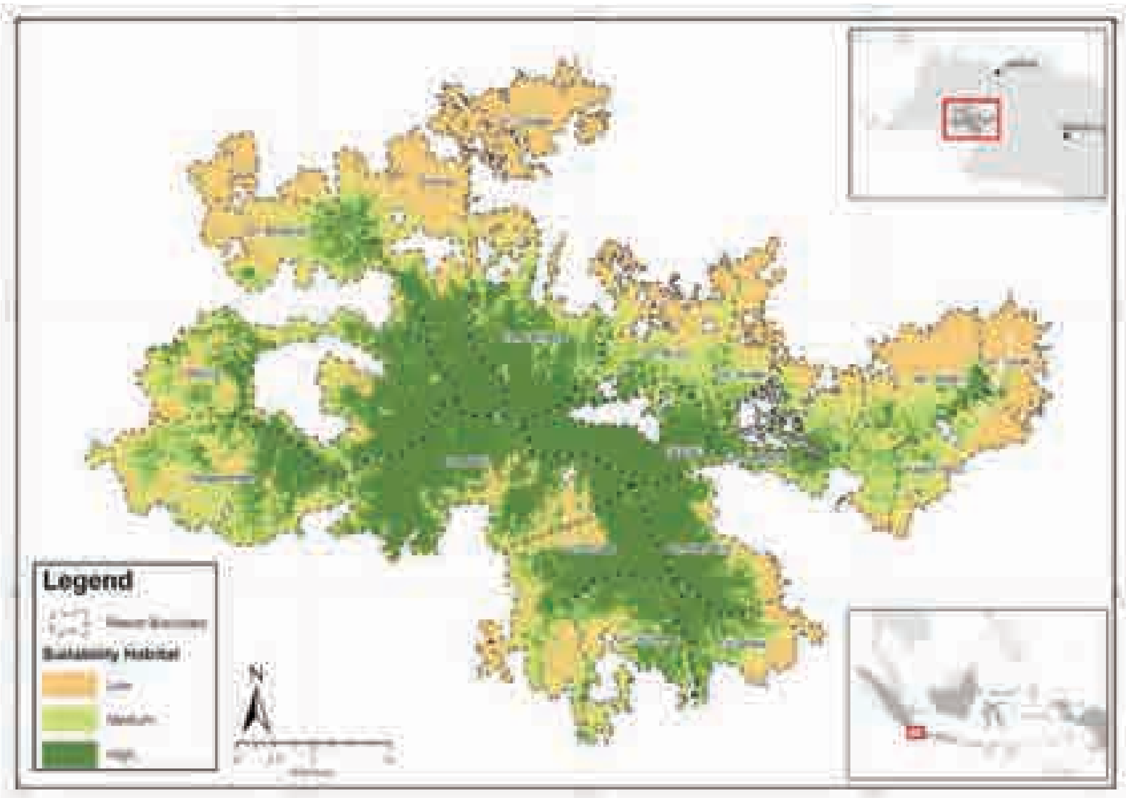

Figure 2. The habitat suitability map for GHSNP indicated that the central part of the park had high suitability while peripheral areas had low suitability for Javan Gibbons.

RESULTS

The final ecological niche model for Javan Gibbons provided a ROC with an AUC of 0.936 for the training data, indicating good performance and suggesting that the model can be used to predict species occurrence. Among the seven environmental variables investigated, forest canopy density and mean annual temperature contributed the most to the model and to predicting Javan gibbon distribution, accounting for 53.9% and 35.6% of the variation in habitat suitability, respectively (Table 2). No other variables in the model were identified as important predictors of habitat suitability for Javan Gibbons.

Most of the area within GHSNP was classified as highly suitable or moderately suitable, with highly suitable habitat comprising the largest proportion of habitat. A total of 36,921 ha (42.1%) of GHSNP was classified as highly suitable habitat, whereas 21,662 ha (24.7%) was classified as moderately suitable, and 29,116 ha (33.2%) was considered to be habitat of low suitability for Javan Gibbons (Figure 2).

DISCUSSION

The MaxEnt analysis confirmed that forest canopy density was the most critical predictor of Javan gibbon distribution in GHSNP and suggested that habitat with dense tree cover is associated with a greater probability of occurrence for this species. Widyastuti et al. (2020) reported similar results for Javan Gibbons in the Dieng Highland in Central Java, where the presence of natural forest with a connected canopy was the most crucial variable predicting habitat suitability in their MaxEnt analysis. Gibbons preferentially use high canopy layers for many activities, including travel, feeding, resting, and singing (Fan et al. 2009; Hamard et al. 2010; Cheyne et al. 2016; Jang et al. 2021). Because Javan Gibbons, like all small apes, primarily travel through brachiation (arm-swinging locomotion that can only be performed across a relatively intact forest canopy), they require high canopy connectivity to travel efficiently and are particularly susceptible to habitat disturbance. High forest connectivity may also indicate high tree density or the presence of large trees, which are associated with the increased availability of plant foods (Zhang et al. 2022) and protection against predators. Our observations suggest that avian predators represent a real threat to gibbons, as we observed the predation attempts from above to the immature individuals by *Spilornis cheela* (Rahayu Oktaviani pers. obs. September 27th, 2019 & February 26th, 2020).

Canopy cover and tree height have also been found to influence the spatial distribution and density of other gibbon species, i.e., Agile Gibbons *Hylobates agilis* (Pang et al. 2022), Borneon White-Bearded Gibbons *Hylobates albibarbis* (Singh et al. 2018), Hoolock Gibbons *Hoolock hoolock* (Alamgir et al. 2015), Yellow Cheeked-Gibbons *Nomascus gabriellae* (Gray et al. 2010), and other arboreal primates, i.e., Borneon Orangutans *Pongo pygmaeus* (Felton et al. 2003), Pied Tamarins *Saguinus bicolor* (Vidal & Cintra 2006), Thomas's Langurs *Presbytis thomasi* (Slater 2015), and Red-Crested Tamarins *Saguinus geoffroyi* (Kim & Riondato 2016).

Climatic conditions have long been observed to play a primary role in limiting species distributions (Gaston 2003; Franklin 2009; Kamilar 2009), either directly or indirectly, through their effects on vegetation (Guisan & Thuiller 2005). Climatic variables may affect the productivity of food plant species that animals consume and, therefore, affect animal behavior, abundance, and distribution (Vidal-García & Serio-Silva 2011). For example, temperature and precipitation affect the distribution of Hoolock Gibbons, likely because of the influence of climate variables on the phenology of fruiting trees (Alamgir et al. 2015; Sarma et al. 2021).

Accordingly, our results showed that mean annual

Table 1. Predictor variables of habitat suitability for Javan Gibbons in GHSNP.

	Environmental variable	Unit	Data source
1	Annual precipitation	Millimeters	Bioclimatic map (http://www.worldclim.org/)
2	Mean annual temperature	°C	Bioclimatic map (http://www.worldclim.org/)
3	Aspect	Degrees	Digital Elevation Model (DEM) SRTM with a 30-meter spatial resolution (http://earthexplorer.usgs.gov/)
4	Distance from river	Meters	The Euclidean distance at software QGIS 2.10
5	Elevation	Meters	Digital Elevation Model (DEM) SRTM with a 30-meter spatial resolution (http://earthexplorer.usgs.gov/)
6	Forest canopy density	%	Imagery 8 2013 using the software Forest Canopy Density Mapper V2
7	Slope	%	Digital Elevation Model (DEM) SRTM with a 30-meter spatial resolution (http://earthexplorer.usgs.gov/)

temperature is the second-most important predictor of Javan Gibbons distribution in GHSNP. This variable is also correlated with elevation, and the relationship with Javan Gibbon distribution may result from an indirect influence of temperature on plant productivity. From an activity budget and behavior perspective, temperature variation may also influence resting time, an essential determinant of primate distribution (Stone et al. 2013; Fei et al. 2019). As a result, feeding and traveling time are generally positively affected by temperature in frugivorous primates (Korstjens et al. 2010; Fan et al. 2012). In future studies, the inclusion of animals experiencing a broader range of ecological conditions could shed more light on Javan Gibbons responses to temperature variation.

The model showed that most of the highly suitable habitat for Javan Gibbons is in the central part of the park, where substantial areas of sub-montane forest have the optimal physical and biotic resources to support Javan Gibbons. However, the area of highly suitable habitat is discontinuous, with some areas fragmented or isolated by areas with lower suitability for Javan Gibbons, especially in the western and eastern parts of the park.

Isolation in habitat fragments could severely threaten Javan Gibbons' long-term survival in these areas. For example, a recent Population and Habitat Viability Analysis for Javan Gibbons in GHSNP by Smith et al. (2017) showed that if the population is fragmented under current pressures, all subpopulations are likely to decline substantially in the next 100 years, and local extinction is very likely for the smallest subpopulations. Thus, maintaining or reestablishing connectivity of fragmented habitats and restoring habitat quality in habitat corridors is critical to facilitating the dispersal of arboreal species like Javan Gibbons across areas of high-quality habitat in GHSNP. Low suitability habitat mainly occurs in the peripheral areas of the park, which may limit Javan Gibbons to more central areas with higher food abundance in GHSNP.

Our species distribution modeling has limitations because it is based on the current realized niche (i.e., it considers where Javan Gibbons occur in the present day) rather than the fundamental niche (the range of places Javan Gibbons could occupy). Other studies have shown that some areas fall under environmental conditions matching the species' ecological environments, although the species does not occur in these areas (Raxworthy et al. 2003; Pearson et al. 2006; Thorn et al. 2009; Abolmaali et al. 2018). The model is also based on surveys at only a limited set of sites within the GHSNP landscape. A more detailed analysis based on a more extensive data set would allow the inclusion of more explanatory variables, which might improve our ability to model the Javan Gibbons ecological niche accurately.

The results of this study add to a growing body of information about Javan Gibbons distribution and habitat suitability in GHSNP, one of the most significant

Image 1. A mountainous survey area in Gunung Halimun Salak National Park.

Image 2. A Javan Gibbon *Hylobates moloch* found during the survey period.

Table 2. Environmental variables and their contribution to habitat suitability in a Maxent model for Javan Gibbons in GHSNP.

	Environmental variable	Predictive value and % contribution
1	Forest canopy density	53.9
2	Annual temperature	35.6
3	Annual precipitation	6.3
4	Slope	2.5
5	Distance from river	1.7
6	Elevation	0.1
7	Aspect	0

remaining habitats for this endangered species (Nijman 2020). The predictive distribution map indicates that suitable habitats for Javan Gibbons are not uniformly distributed across GHSNP; some areas in GHSNP are more suitable than others for the species. Most of the suitable area is in the central part of the park, which must be protected to optimize the habitat and ensure the long-term persistence of the species. In addition, some high-quality habitat is located in peripheral areas of GHSNP. To prevent further degradation of these areas and to maintain and improve connectivity between fragments of high-quality habitat, buffer areas surrounding areas of high-quality habitat should be protected and, where possible, restored.

To ensure the long-term persistence of Javan Gibbons, an endangered species endemic to Indonesia, we recommend that the Indonesian Ministry of Environment and Forestry (MoEF) and the GHSNP authorities prioritize habitat protection to prevent erosion and degradation of high-quality habitats, including the area of Resort Cikaniki, Gunung Kendeng, and Gunung Bedil. Habitat restoration to increase habitat quality in degraded habitat in the peripheral areas of the park (i.e., the area of Resort Gunung Bongkok, Cisoka, and Gunung Talaga) is crucial to improve the low-medium suitable habitat adjacent to higher-quality habitat patches, especially in the corridor area connected the region of Halimun and Mount Salak as part of their conservation management strategy.

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Indonesian abstract: Pemodelan distribusi spesies menjadi sebuah alat penting untuk memahami ekologi suatu spesies dan telah banyak diaplikasikan dalam bidang konservasi. Melalui pemodelan Maximum Entropy (Maxent), kami mengidentifikasi faktor-faktor kunci untuk menentukan sebaran potensial bagi Owa Jawa *Hylobates moloch* yang terancam punah di salah satu habitat tersisa di Taman Nasional Gunung Halimun Salak (TNGHS), Indonesia, antara bulan Oktober dan November 2015, serta bulan April dan Mei 2016. Hasil analisis Maxent menunjukkan bahwa kerapatan tajuk pohon dan suhu menjadi faktor utama dalam memprediksi sebaran Owa Jawa. Kedua faktor utama tersebut memiliki skor kontribusi masing-masing sebesar 53.9% dan 35.6%. Sementara, peta prediksi sebaran Owa Jawa menunjukkan bahwa habitat yang sesuai tersebar secara tidak merata di dalam kawasan TNGHS. Habitat yang memiliki kesesuaian tinggi memiliki proporsi terbesar, dimana 42.1% kawasan TNGHS diklasifikasikan sebagai habitat dengan tingkat kesesuaian tinggi, sedangkan 24.7% dari total luas kawasan memiliki tingkat kesesuaian sedang, dan 33.2% merupakan habitat dengan kesesuaian rendah. Prioritas pengelolaan kawasan harus difokuskan untuk meningkatkan kualitas habitat di kawasan terdegradasi, serta perlu dilakukan patroli rutin dan penegakan hukum untuk mengurangi kerusakan habitat di Kawasan TNGHS.

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Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23472–23486

OPEN ACCESS

ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)

https://doi.org/10.11609/jott.8467.15.7.23472-23486

#8467 | Received 02 April 2023 | Final received 31 May 2023 | Finally accepted 18 July 2023

ARTICLE

Babesa Sewage Treatment Plant as a vital artificial wetland habitat for a multitude of avian species

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Abstract. This study aimed to glean basic ecological aspects on diversity and abundance, temporal variation and guild composition of the birds at Babesa Sewage Treatment Plant (STP). The line transect method was used as the sampling technique from November 2021 to October 2022. A total of 80 species belonging to 58 genera, 29 families, and 11 orders were detected, of which three, namely, River Lapwing *Vanellus duvaucelii*, Falcated Duck *Mareca falcata*, and Ferruginous Duck *Aythya nyroca*, are 'Near Threatened' with the remaining being 'Least Concern'. The highest species richness was recorded in the winter (6.29), the highest species diversity in the spring (2.73), and the highest evenness in the summer (0.76). There was not any statistically significant difference between non-waterbirds and waterbirds, or between feeding guilds. However, based on a permutational multivariate analysis of variance (PERMANOVA), the bird composition was significantly different among seasons. Subsequently, pairwise comparisons revealed a significant difference between autumn & winter (P = 0.006), autumn & summer (P = 0.006), autumn & spring (P = 0.018), winter & summer (P = 0.006), winter & spring (P = 0.006) as well as spring & summer (P = 0.006). The non-metric multidimensional scaling (NMDS) biplot showed most bird species overlap occurred between autumn and spring as well as summer and spring, respectively. Taken together, the present results suggest that the Babesa STP holds significant potential as a habitat for diverse avian populations and underscores the ecological significance of artificial wetlands.

Keywords: Artificial wetland, avian population, feeding guilds, non-waterbirds, species diversity, waterbirds.

Editor: H. Byju, Coimbatore, Tamil Nadu, India.

Date of publication: 26 July 2023 (online & print)

Citation: Nima, P., M. Timsina, T. Jamtsho & P. Khandu (2023). Babesa Sewage Treatment Plant as a vital artificial wetland habitat for a multitude of avian species. *Journal of Threatened Taxa* 15(7): 23472–23486. https://doi.org/10.11609/jott.8467.15.7.23472-23486

Funding: This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Competing interests: The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Author contributions: PN conceived the study design, carried out field study, data collection, data tabulation, statistical analysis and prepared the whole manuscript. MT carried out field study, data collection, data tabulation and helped in revision. TJ provided critical comments on the whole manuscripts and helped in revision. PK critically commented and revised the whole manuscript, and provided comments on statistical analysis. All authors read and approved the final manuscript.

Acknowledgements: All authors remain indebted to IDEA WILD for the grant of the equipment (Police Binocular).

INTRODUCTION

Accumulating evidence suggest that wetlands are indispensable for the conservation of many waterbirds and migratory species as well as for mammals, fishes, invertebrates, reptiles and amphibians (Airoldi et al. 2008; Kedleck & Wallace 2008; Engle 2011). This is because wetlands are primarily considered to be abundant in food (Rajpar et al. 2010) and water resources that sustain various lifeforms. Particularly for waterbirds, they are thought to provide breeding, stopover and wintering sites for diverse migratory species (Rendon 2008; Ma et al. 2009), and have been shown to help in the accumulation of critical energy reserves (Catry et al. 2022; Liu et al. 2022), which is inevitable for the wetland-dependent birds to complete a long migration (Alerstam et al. 2003). Wetlands are also considered to enhance landscape biodiversity, control floods, provide recreation (Hansson et al. 2005) and remove pollutants (Vymazal 2010).

However, due to the burgeoning human population, wetlands have been imperilled (Zedler & Kercher 2005). For example, anthropogenic-induced pressures such as water pollution, surplus use of pesticides in adjoining agricultural habitats and human settlements have caused 50% of natural wetlands to be degraded and altered globally (Mitsch & Gosselink 2015). Likewise, human dependence on wetlands for various ecosystem services has intensified and mounted pressures on these ecologically delicate ecosystems (Molur et al. 2011), which may further deteriorate in the future.

Consequently, it has placed wetland inhabitants in a perilous state (Soderquist et al. 2021) often culminating in fewer resources for wetland-dependent species such as waterbirds (Forcey et al. 2011). As a result, avifaunal diversity has diminished. Thus, waterbirds have become progressively reliant on alternative and artificial wetlands (Murray & Hamilton 2010) such as small agricultural ponds, paddy fields and water treatment plants to meet their needs (Lawler 2001; Sebastián-González et al. 2010; Hsu et al. 2011).

Though artificial wetlands cannot fully replace the operationality of natural wetland habitats (Li et al. 2013), wastewater treatment ponds have been reported to increasingly play an important role in supporting regional population of waterbirds (Kalejta-Summers et al. 2001) mainly due to abundance of food resources such as zooplankton (Hamilton et al. 2005). Further, such artificial wetland habitats have been reported to form key staging sites and breeding grounds for migratory bird species (Donahue 2006). Indeed, Breed et al. (2020) showed that wastewater treatment plant is a crucial refuge site for several species of ducks and waders. Similarly, several other studies have also shown that sewage treatment plant (STP) provide habitat supplements and occasional alternative sanctuaries for waterbirds (Attuquayefio & Gbogbo 2001; Gbogbo 2007; Harebottle et al. 2008; Murray & Hamilton 2010). As a consequence, attempts have been made globally to safeguard the wetlands of significance (Tiéga 2011; Ibrahim & Aziz 2012), several of which encompass artificial wetlands (Zedler & Kercher 2005). For instance, a few sewerage habitats, such as Phakalane sewage lagoons in Botswana and Samra sewage in Jordan, are internationally acknowledged as an important bird area (Orlowski 2013).

However, despite the global recognition of STPs as valuable habitat for many bird species, studies pertaining to it are limited (Murray & Hamilton 2010). As such, there is not a single report from Bhutan regarding the role of STP in bird conservation, and in general, studies concerning bird diversity and conservation are sparse and limited only to protected areas (Gyeltshen et al. 2020; Dendup et al. 2021), non-protected areas (Norbu et al. 2021) and freshwater ecosystems (Passang 2018; Nima & Dorji 2022). Therefore, there is a paucity of information and a knowledge gap concerning the role of STPs on the conservation of waterbirds in Bhutan.

To this end, the present systematic study aimed to glean basic ecological aspects on i) diversity and abundance, ii) temporal variation and iii) guild composition of the birds found in Thimphu's only STP. This study will also provide the opportunity to form a basis for formulating national and local policies for the conservation of waterbird species (Wang et al. 2018) and proper management of their essential habitats such as the STP. Documenting the avian diversity of this habitat will advance our understanding of the utilization of sewerage treatment plants by the different avian communities.

MATERIALS AND METHODS

Study site

The present study was conducted at Babesa STP (27.4367^{0}N, 89.6521^{0}E) (Figure 1), Thimphu, Bhutan. The study site spans an area of 13 acres of land with the design capacity of 1.75 million l/day and 325 mg/l five-day biological oxygen demand (BOD_5) removal (Phuntsho et al. 2016). There are three ponds with varying areas and depth. The first one, anaerobic pond covers 1.85

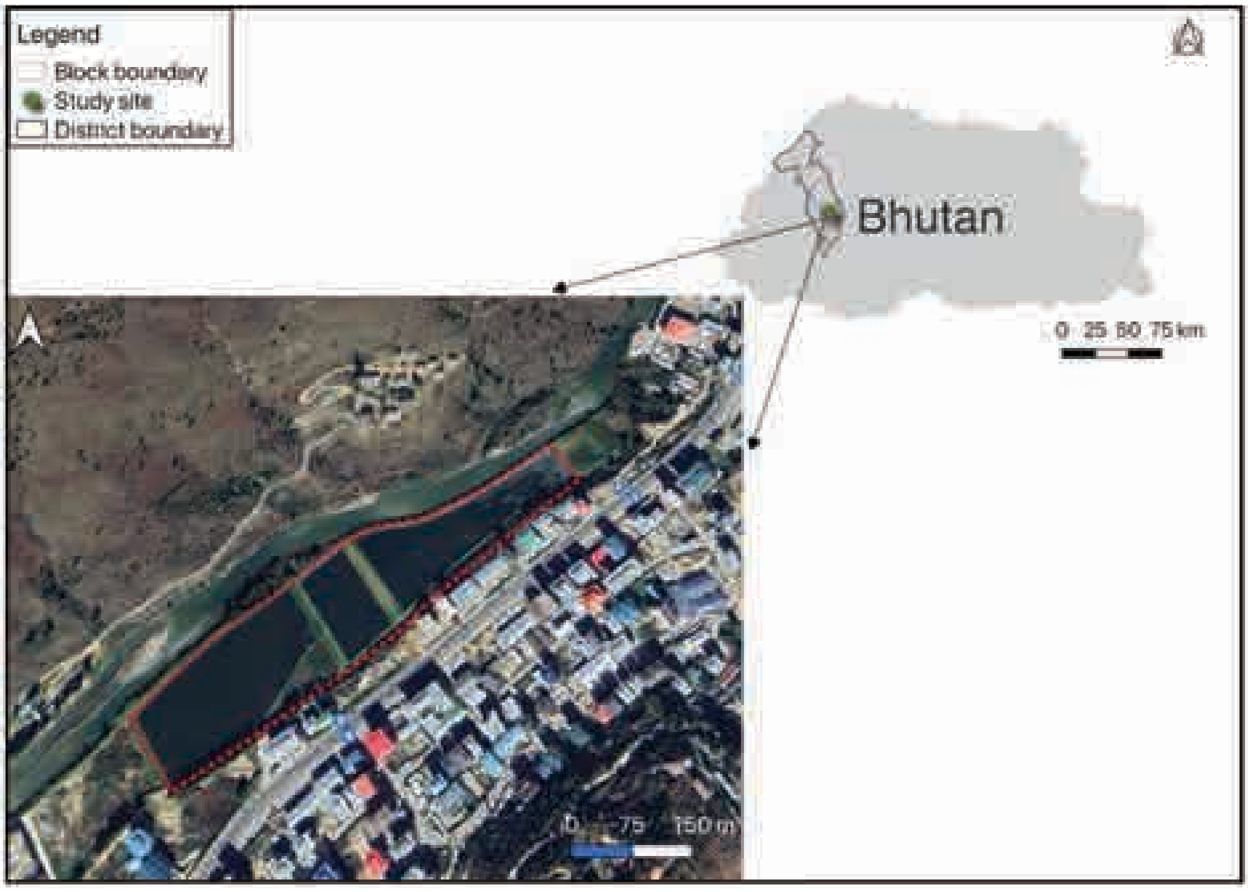

Figure 1. Map showing the location of the study site. The boundary of the STP is marked with red dotted lines.

ha with a depth of 3 m, while the second, facultative pond covers 0.71 ha with a depth of 2 m, and the third, maturation pond covers 1.71 ha with a depth of 1.5 m, respectively. The banks of all the three ponds have flat upper surfaces lined with rocks, mostly covered by *Cynodont dactylon,* and features steep vertical slopes approximately measuring 0.45 m. Other sparsely populated herb species such as *Rumex nepalensis* is also found along the edges of the pond. The surrounding vegetation is mostly dominated by tree species such as *Salix babylonica*, sparsely populated *Silax* and *Populus* species along with the shrub *Rosa brunonii* and the herb *Fagopyrum* species.

It is situated about 40 m away from Babesa-Thimphu expressway and lies to the immediate south of Wangchhu (chhu = river) while heading towards the main town. The nearest human settlement is about 15 m away from the study site. The site has moderate summer, cool spring and autumn, and a cold winter season with an annual average temperature of 13.8°C, and an annual average rainfall of 48.3 mm (NCHM 2013). The STP uses wastewater stabilization ponds alone (Phuntsho et al. 2016).

Bird counts

A reconnaissance study was carried out in the last week of October 2021 to identify vantage points and a suitable position for a transect lines. The actual study commenced from the first week of November 2021, considered to be the ideal time for studying wintering and resident birds in the sub-Himalayan region (Salewski et al. 2003; Mazumdar et al. 2007), through to the end of October 2022.

We divided the time of the day into two intervals: 0800–1000 h in the morning and 1500–1700 h in the evening for 23 bird count surveys along the 650-m transect line. So, in a day we traversed for four hours along the 1,300 m transect line. For the remaining 14 bird surveys, in a day we surveyed the birds only once for 2 h in the evening along the 650 m transect line. Altogether, we spent 120 h surveying the birds along the 39,000 m of transect line. All the surveys were performed on weekends.

Prior to entering the designated study site, we observed and recorded all the birds sighted in the open sewerage pond from a vantage point to make a quick estimate of the actual birds present and help validate the counts made from the line transect. Before we traversed the preset transect line by foot and recorded the sightings, we spent about 10 min to settle so that the birds did not feel disturbed and stressed. Concurrently, care was taken to maintain a proper distance between the observer and birds. At a certain randomly identified points marked along the transect, we stopped for about 15 min and recorded additional visible species and estimated the number of each species (Webb et al. 2010). We included all the observed bird species either wandering on the bank or resting on the bank or trees as long as they were within 50 m radius from the transect (Hutto et al. 1986). We did not consider flying birds in order to avoid repeated counting of the same individuals. Moreover, to reduce the impact of inclement weather on results of sightings, observations were not taken during snowfall or rainfall.

Birds were recorded using direct observations with the help of binoculars namely Police (7 x 50, Steiner, Germany), and Nikon (7 x 50), and immediately noted in the field journal. Where a bird species could not be confirmed, photos were taken using Canon 7d Mark II paired with Tamron G2 telephoto zoom lens (150–600 mm) and Nikon D850 paired with Nikkor telephoto zoom lens (200–500 mm) for further identification.

Bird identification, nomenclature, feeding guild, and conservation status

We followed Grimmett et al. (2019) for avifauna identification and nomenclature. Further, birds were categorized as per their residency pattern as Altitudinal Migrant (AM), Passage Migrant (PM), Resident (R), Summer Visitor (SV), Vagrant (V), and Winter Visitor (WV) (Ali et al. 1996; Feijen & Feijen 2008; Grimmett et al. 2019). Likewise, feeding guilds were ascribed based on the observation made in the field (Kumar & Sharma 2018; Singh et al. 2020). Additionally, we followed Ali & Ripley (1987) to assign the feeding guild: granivorous if they fed on grains, omnivorous if they fed on both plants and animals, insectivorous if they fed on insects, carnivorous if they fed on non-insects' invertebrates and vertebrates, frugivorous if they fed on fruits and nectarivorous if they fed on floral nectar. Birds were also categorized as water and non-waterbirds. The conservation status of the identified bird species was categorized as per International Union of Conservation for Nature (IUCN 2022).

Species accumulation curve

Species accumulation curve as a function of sampling adequacy was performed to determine if the probability of sighting new species increased with increase in sampling days. The function 'specaccum' from R package 'vegan' (Oksanen et al. 2019) was employed to discover the expected species accumulation curve by means of sample-based rarefaction (Chiarucci et al. 2008).

Bird abundance and rank abundance curve

We followed Bull (1974) to describe the bird abundance. If more than 1,000 individuals were seen in a day, it was classed as very abundant (VA), those between 201–1,000 individuals as abundant (A), between 51–200 individuals as very common (VC) and those between 21–50 as common (C). Likewise, those between seven to 20 were classed as fairly common (FC) and between one to six as uncommon (UC). For birds with one to six individuals per season, it was classed as rare (Ra) and those with infrequent occurrence as very rare (VR) species.

The season-wise rank abundance curve was graphed with abundance rank and relative abundance. For interpretation purpose, a horizontal rank abundance indicated a community with a complete even distribution, whereas a steeper slope indicated a community with a less even distribution of species (Akinnifesi 2010). Subsequently, a rank abundance curve was plotted to analyse dominance patterns and species evenness across different seasons.

Data analysis

The relative diversity (RDi) of families was computed following La Torre-Cuadros et al. (2007), where:

$$\textbf{RDi} = \frac{\text{Number of species in a family}}{\text{Total number of species}} * 100.$$

For species evenness (E), we followed Pielou's index (Pielou 1966):

$$E = \frac{H'}{\text{LnS}}$$

Where:

E: Pielou's index

H': Shannon diversity index

Ln: natural logarithm

S: number of species observed

If E is close to 0, species evenness is considered low and if E is close to 1, evenness is considered to be relatively uniform.

For richness index (R), we followed Margalef's equation (Margalef 1968):

$$R = \frac{(S-1)(1)}{\text{Ln}(N)}$$

Where:

R: index of species richness.

S: number of species observed.

N: number of individuals of all species observed.

Ln: natural logarithm.

If R <2.5, the species richness is considered low, medium if R >2.5 but <4 and high if R >4.

For species diversity, Shannon-Weaver index (H’) (Shannon & Weaver 1949) was used as follows:

$$\text{Shannon - Weaver index (H')} = -\sum_{i=1}^{n} Pi \ln Pi$$

Where:

H’: Shannon-Weaver diversity index.

n: number of individual species.

Pi: proportion of individual species belonging to the i^{th} species of the total number of individuals.

If H’ <1, the diversity index is considered low, medium if H’ >1 but <3 and high if H’ >4.

Data was checked for normality using Shapiro-Wilk test. As it did not conform to a normal distribution, a non-parametric Kruskal-Wallis test was performed to evaluate the statistical significance in the feeding guilds of the birds. Likewise, to assess the statistical significance between waterbirds and non-waterbirds, a Mann-Whitney test was computed. Waterbirds included Anatidae, Ardeidae, Charadriidae, Cinclidae, Ibidorhynchidae, Motacillidae (White Wagtail *Motacilla alba*, White-browed Wagtail *Motacilla maderaspatensis*, Water Pipit *Anthus spinoletta*, Citrine Wagtail *Motacilla citreola*), Muscicapidae (White-capped Redstart *Phoenicurus leucocephalus*, Plumbeous Redstart *Phoenicurus fuliginosus*), Podicipedidae, Phalacrocoracidae, Rallidae, and Scolopacidae.

NMDS was applied to visualize and compare species composition across seasons using the function ‘ordihull’ in vegan (Tojo 2015) and the results were presented as two-dimensional plots. The function ‘ordihull’ creates neat and convex outlines to further depict group segregation for visual clarity (Moskowitz et al. 2020).

We removed species whose frequency of observation was only once. NMDS is an ordination technique that uses rank-order dissimilarity of multivariate data to ordinate sites and species, in which similar communities are placed closer together (Duchardt et al. 2018). To this end, we used Bray-Curtis dissimilarity, which factors in species abundance, using vegan package (Bray & Curtis 1957).

The statistical difference in species composition across seasons was computed by PERMANOVA using ‘adonis’ function from the vegan package (Oksanen et al. 2020). Subsequently, to evaluate which seasons significantly differed from each other, pairwise ‘adonis’ function in R with Bonferroni correction was used (Arbizu 2020). Abundance values were square root-transformed to lower the influence of abundant species on rare species prior to executing multivariate analysis method (Zar 2010).

All analyses were performed by using R Statistical Computing Software, version 4.0.2. P <0.05 was considered statistically significant for all analyses.

RESULTS

Sampling adequacy and Species composition

Sampling adequacy was tested based on the number of bird species sighted during the study period, which indicated that an asymptote was not reached. Hence, it is plausible that a greater number of unrecorded bird species might be present at the site (Figure 2).

During a period spanning from November 2021 to October 2022, the present study recorded a total of 7661 individual birds belonging to 80 species, 58 genera, 29 families and 11 orders (Table 1). The greatest number of bird species detected were from order Passeriformes (52.50%) with 42 species, followed by Anseriformes (18.75%) with 15 species, Charadriiformes (7.5%) with six species, Gruiformes (5%) with four species, Pelecaniformes (3.75%) with three species, Accipitriformes, Columbiformes, Coraciiformes, Podicipediformes with two species (2.50%) each, and Bucerotiformes and Suliformes with only one species (1.25%) each.

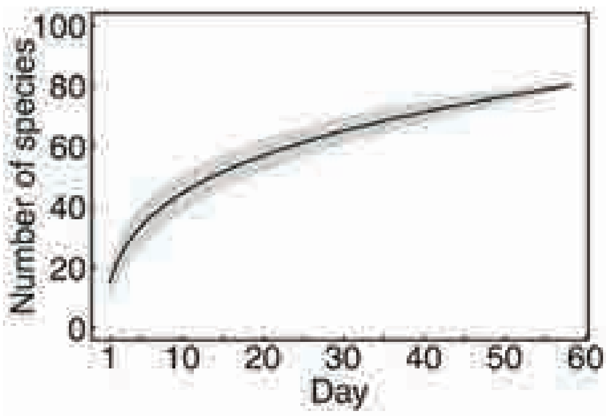

Figure 2. Species accumulation as a function of number of sampling days. The grey shade indicates the 95% confidence interval.

Table 1. Family, order and species recorded from November 2021 to October 2022 from the study site.

Family	Order	Common name	Scientific name
Muscicapidae	Passeriformes Passeriformes Passeriformes Passeriformes Passeriformes Passeriformes Passeriformes Passeriformes	Plumbeous Redstart Hodgson's Redstart Aberrant Bush-warbler White-capped Redstart Slaty-backed Flycatcher Common Stonechat Chestnut-bellied Rock-Thrush Verditer Flycatcher	*Phoenicurus fuliginosus* *Phoenicurus hodgsoni* *Horornis flavolivaceus* *Phoenicurus leucocephalus* *Ficedula erithacus* *Saxicola maurus* *Monticola rufiventris* *Eumyias thalassinus*
Motacillidae	Passeriformes Passeriformes Passeriformes Passeriformes Passeriformes Passeriformes Passeriformes	White Wagtail Olive-backed Pipit White-browed Wagtail Grey Wagtail Water Pipit Rosy Pipit Citrine Wagtail	*Motacilla alba* *Anthus hodgsoni* *Motacilla maderaspatensis* *Motacilla cinerea* *Anthus spinoletta* *Anthus roseatus* *Motacilla citreola*
Leiothrichidae	Passeriformes Passeriformes Passeriformes Passeriformes	Chestnut-crowned Laughingthrush Rufous Sibia Red-billed Leiothrix Chestnut-tailed Minla	*Trochalopteron erythrocephalum* *Heterophasia capistrata* *Leiothrix lutea* *Chrysominla strigula*
Corvidae	Passeriformes Passeriformes	Large-billed Crow House Crow	*Corvus macrorhynchos* *Corvus splendens*
Turdidae	Passeriformes Passeriformes Passeriformes Passeriformes Passeriformes	Blue Whistling-thrush Black-throated Thrush Alpine Thrush White-collared Blackbird Red-throated Thrush	*Myophonus caeruleus* *Turdus atrogularis* *Zoothera mollissima* *Turdus albocinctus* *Turdus ruficollis*
Zosteropidae	Passeriformes Passeriformes	Indian White-eye Whiskered Yuhina	*Zosterops palpebrosus* *Yuhina flavicollis*
Paridae	Passeriformes Passeriformes	Green-backed Tit Coal Tit	*Parus monticolus* *Periparus ater*
Passeridae	Passeriformes Passeriformes	Eurasian Tree Sparrow Russet Sparrow	*Passer montanus* *Passer cinnamomeus*
Phylloscopidae	Passeriformes Passeriformes	Common Chiffchaff Sulphur-bellied Warbler	*Phylloscopus collybita* *Phylloscopus griseolus*
Pycnonotidae	Passeriformes	Himalayan Black Bulbul	*Hypsipetes leucocephalus*
Aegithalidae	Passeriformes	Rufous-fronted Bushtit	*Aegithalos iouschistos*
Cettiidae	Passeriformes	Aberrant Bush Warbler	*Horornis flavolivaceus*
Emberizidae	Passeriformes	Little Bunting	*Emberiza pusilla*
Fringillidae	Passeriformes	Yellow-breasted Greenfinch	*Chloris spinoides*
Cinclidae	Passeriformes	Brown Dipper	*Cinclus pallasii*
Laniidae	Passeriformes	Grey-backed Shrike	*Lanius tephronotus*
Prunellidae	Passeriformes	Rufous-breasted Accentor	*Prunella strophiata*
Anatidae	Anseriformes Anseriformes Anseriformes Anseriformes Anseriformes Anseriformes Anseriformes Anseriformes Anseriformes Anseriformes Anseriformes Anseriformes Anseriformes Anseriformes Anseriformes	Ruddy Shelduck Common Shelduck Common Merganser Mallard Red-crested Pochard Eastern Spot-billed Duck Common Teal Falcated Duck Northern Pintail Northern Shoveler Gadwall Eurasian Wigeon Ferruginous Duck Tufted Duck Garganey	*Tadorna ferruginea* *Tadorna tadorna* *Mergus merganser* *Anas platyrhynchos* *Netta rufina* *Anas zonorhyncha* *Anas crecca* *Mareca falcata* *Anas acuta* *Spatula clypeata* *Mareca Strepera* *Mareca penelope* *Aythya nyroca* *Aythya fuligula* *Spatula querquedula*
Alcedinidae	Coraciiformes Coraciiformes	Crested Kingfisher Common Kingfisher	*Megaceryle lugubris* *Alcedo atthis*
Charadriidae	Charadriiformes Charadriiformes Charadriiformes	River Lapwing Long-billed Plover Grey-headed Lapwing	*Vanellus duvaucelii* *Charadrius placidus* *Vanellus cinereus*
Scolopacidae	Charadriiformes Charadriiformes	Common Sandpiper Green Sandpiper	*Actitis hypoleucos* *Tringa ochropus*

Family	Order	Common name	Scientific name
Ibidorhynchidae	Charadriiformes	Ibisbill	*Ibidorhyncha struthersii*
Columbidae	Columbiformes Columbiformes	Oriental Turtle-dove Rock Pigeon	*Streptopelia orientalis* *Columba livia*
Accipitridae	Accipitriformes Accipitriformes	Long-legged Buzzard Himalayan Buzzard	*Buteo rufinus* *Buteo refectus*
Rallidae	Gruiformes Gruiformes Gruiformes Gruiformes	Eurasian Coot Eurasian Moorhen White-breasted Waterhen Black-tailed Crake	*Fulica atra* *Gallinula chloropus* *Amaurornis phoenicurus* *Zapornia bicolor*
Ardeidae	Pelecaniformes Pelecaniformes Pelecaniformes	Indian Pond-Heron Cattle Egret Little Egret	*Ardeola grayii* *Bubulcus ibis* *Egretta garzetta*
Podicipedidae	Podicipediformes Podicipediformes	Black-necked Grebe Great Crested Grebe	*Podiceps nigricollis* *Podiceps cristatus*
Phalacrocoracidae	Suliformes	Great Cormorant	*Phalacrocorax carbo*
Upupidae	Bucerotiformes	Common Hoopoe	*Upupa epops*

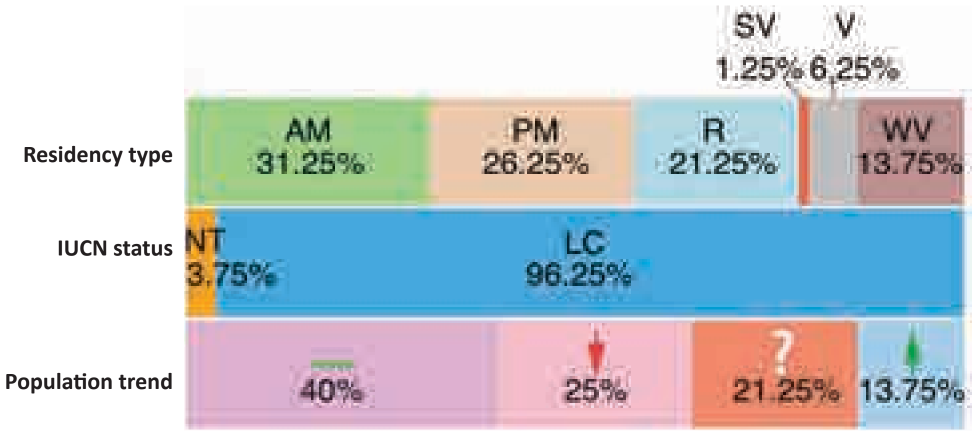

Figure 3. Graph showing residential status, IUCN status and population trend of the species in percentage: AM—Altitudinal Migrant | PM—Passage Migrant | R—Resident | SV—Summer Visitor | V—Vagrant | WV—Winter visitor | NT—Near Threatened | LC—Least Concern. — -Stable | ↓—Decreasing| ?—Unknown| ↑—Increasing.

Global population trends and residential status

Of the 80 recorded bird species, only three birds namely River Lapwing, Falcated Duck, and Ferruginous Duck were 'Near Threatened' species classified based on the IUCN Red List category. The remaining birds were species of 'Least Concern'. Further, the present study found out that sewerage treatment plant hosted 32 species (40%) of birds known to have a stable population trend, 11 increasing (13.75%), 20 decreasing (25%) and 17 (21.25%) unknown on the global population trends as per the IUCN. The study also recorded the residential status of the birds and found 31.25% (AM), 26.25% (PM), 21.25% (R), 1.25% (SV), 6.25% (V), and 13.75% (WV), respectively (Figure 3).

Relative diversity, Bird abundance, and Rank abundance

Table 2 shows the relative diversity of the bird families. Subsequently, Anatidae (15 species, RDi = 18.75) was found to be the most dominant of the total 29 families followed by Muscicapidae (eight species, RDi = 10), Motacillidae (seven species, RDi = 8.75), Turdidae (five species, RDi = 6.25), Leiothrichidae and Rallidae (four species each, RDi = 5), Ardeidae and Charadriidae (three species each, RDi = 3.75), Accipitridae, Alcedinidae, Columbidae, Corvidae, Paridae, Passeridae, Phylloscopidae, Podicipedidae, Scolopacidae and Zosteropidae (two species each, RDi = 2.50). The poorly represented families were Ibidorhynchidae, Aegithalidae, Cettiidae, Cinclidae, Emberizidae, Fringillidae, Laniidae, Phalacrocoracidae, Prunellidae, Pycnonotidae and Upupidae (one species each, RDi =

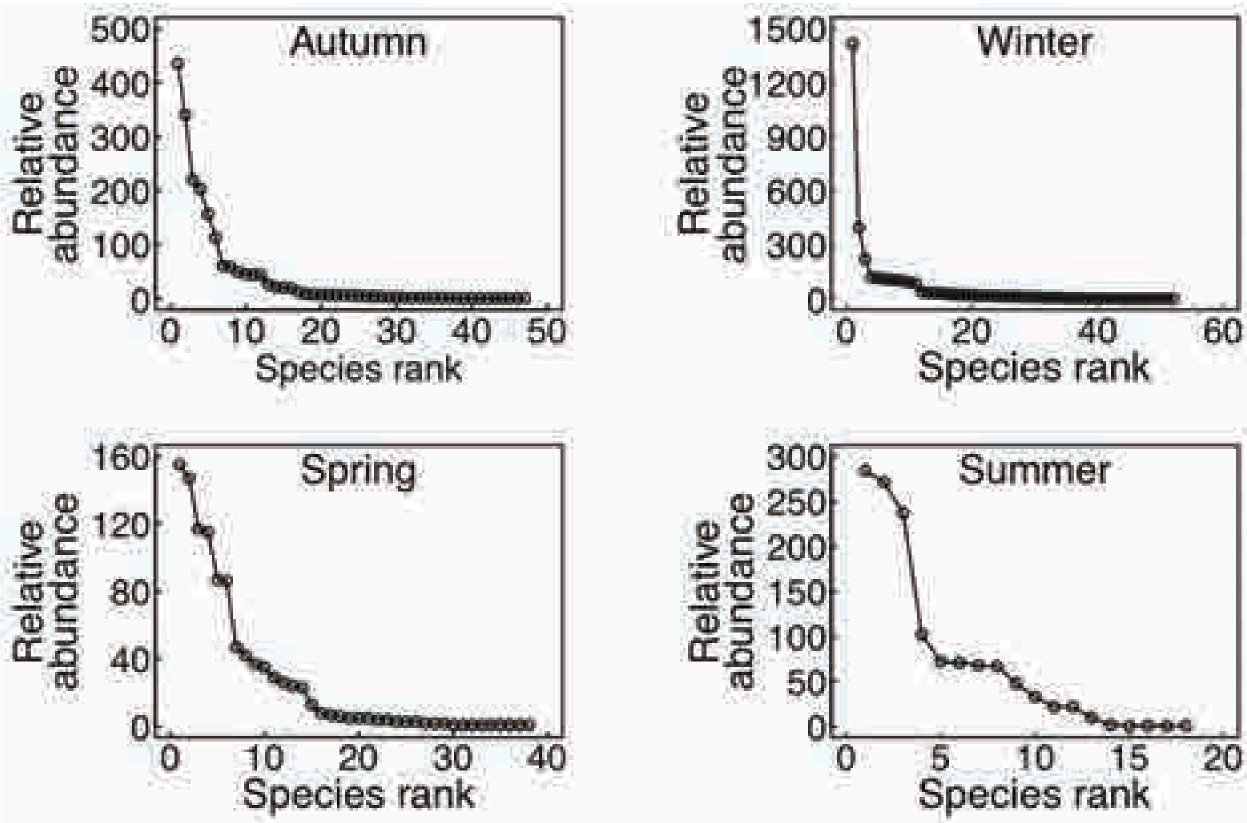

Figure 4. Rank abundance curve for bird species in autumn, winter, spring and summer.

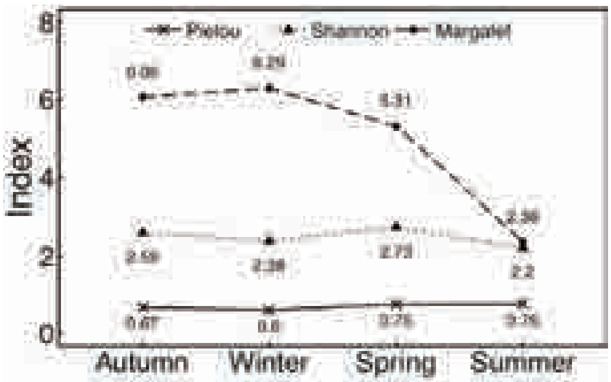

Figure 5. Seasonal variation in Pielou's evenness, Shannon-Weaver diversity index and Margalef's richness index.

1.25). Assessment of the bird abundance showed that three species were VC, eight species (C), 12 species (FC), eight species (UC), 13 (Ra) and 36 species (VR).

The rank-abundance curve had a steep gradient for winter, autumn and spring season, respectively, denoting low evenness of bird species (Figure 4). During winter, Ruddy Shelduck *Tadorna ferruginea* ranked first followed by White Wagtail, Common Merganser *Mergus merganser*, Common Sandpiper *Actitis hypoleucos*, and River Lapwing *Vanellus duvaucelii*. In the autumn season, White Wagtail ranked first followed by Ruddy Shelduck, Oriental Turtle-Dove *Streptopelia orientalis*, River Lapwing, and Common Sandpiper. Spring season had White Wagtail ranked first followed by River Lapwing, Oriental Turtle-Dove, House Crow *Corvus splendens* and Common Sandpiper. By contrast, the curve for summer season was shallower in comparison to the other seasons. Subsequently, summer witnessed higher even distribution of the birds with Oriental Turtle-dove ranked first followed by River Lapwing, White Wagtail, Himalayan Black Bulbul *Hypsipetes leucocephalus* and Eurasian Hoopoe *Upupa epops*. Moreover, the curve length of summer and autumn season are shorter compared to the winter and spring season.

Richness index and Species diversity

Figure 5 shows season-wise Margalef's richness index (R), Shannon-Weaver diversity index (H') and Pileou's evenness index. Winter had the highest species richness (6.29), followed by autumn (6.06), spring (5.31) and summer (2.36), respectively. Similarly, the highest species diversity was recorded for the spring season (2.73), followed by autumn (2.59), winter (2.38) and summer (2.20), respectively. The highest evenness was recorded for summer (0.76), followed by spring (0.75), autumn (0.67) and winter (0.60), respectively.

Feeding guilds of birds and difference between waterbirds and non-waterbirds

Figure 6 shows the abundance of birds in different feeding guilds. A non-parametric Kruskal-Wallis test was carried out to check for statistically significant difference between the guilds. Result revealed that there was no statistically significant difference between the feeding guilds (x^2 = 2.14, df = 3, P = 0.543). However, insectivores were higher (median = 17.0, Q1–Q3 = 1.0–45.0) than granivores (median = 12.0, Q1–Q3 = 8.5–126.5), omnivores (median = 8.5, Q1–Q3 = 1.0–40.25) and carnivores (median = 4.0, Q1–Q3 = 1–7.00).

Likewise, Figure 7 shows the relative abundance of waterbirds and non-waterbirds. A Mann-Whitney test found that there was no statistically significant difference between the relative abundance of waterbirds and non-waterbirds (Z = -0.2769, P = 0.78), although non waterbirds were higher (median = 10.0, Q1 – Q3 = 1–42.50) than the waterbirds (median = 7.0, Q1–Q3 = 2–41.0).

Comparisons of bird species composition across seasons

The NMDS analysis revealed a stress value of 0.146 and suggested a good fit (Clarke & Warwick 2001). The NMDS biplot showed that most bird species overlap occurred between autumn and spring seasons as well as summer and spring, respectively. However, the overlap did not occur between winter and spring, winter and summer as well as between autumn and summer (Figure 8).

To check for statistically significant difference in the bird species composition across seasons, a PERMANOVA test was computed and found that there was a statistically significant difference ($F_{3, 56}$ =16.732, P = 0.001).

Subsequently, pairwise comparisons revealed a statistically significant difference between autumn and winter (R^2 = 0.347, P = 0.006, df = 1), autumn and summer (R^2 = 0.242, P = 0.006, df = 1), autumn and spring (R^2 = 0.148, P = 0.018, df = 1), winter and summer (R^2 = 0.706, P = 0.006, df = 1), winter and spring (R^2 = 0.502, P = 0.006, df = 1) as well as spring and summer (R^2 = 0.197, P = 0.006, df = 1), respectively.

Table 2. The number of species in each avian family and their relative diversity.

Avian families	Number of species	Relative diversity (RDi)
Accipitridae	2	2.50
Aegithalidae	1	1.25
Alcedinidae	2	2.50
Anatidae	15	18.75
Ardeidae	3	3.75
Cettiidae	1	1.25
Charadriidae	3	3.75
Cinclidae	1	1.25
Columbidae	2	2.50
Corvidae	2	2.50
Emberizidae	1	1.25
Fringillidae	1	1.25
Ibidorhynchidae	1	1.25
Laniidae	1	1.25
Leiothrichidae	4	5.00
Motacillidae	7	8.75
Muscicapidae	8	10.00
Paridae	2	2.50
Passeridae	2	2.50
Phalacrocoracidae	1	1.25
Phylloscopidae	2	2.50
Podicipedidae	2	2.50
Prunellidae	1	1.25
Pycnonotidae	1	1.25
Rallidae	4	5.00
Scolopacidae	2	2.50
Turdidae	5	6.25
Upupidae	1	1.25
Zosteropidae	2	2.50

DISCUSSION

To our knowledge, this is the first study that reported on the avifaunal composition concerning species diversity, relative abundance, feeding guilds and temporal variation from the Babesa STP, Bhutan. Despite the rapid urban sprawl over the years, a substantial number of avian species was observed at the study site.

In total, 80 species of birds, representing about 12.05% of the country's total bird species, belonging to 58 genera, 29 families and 11 orders were detected accounting for a total of 7661 individuals. The most notable and the relatively abundant bird species

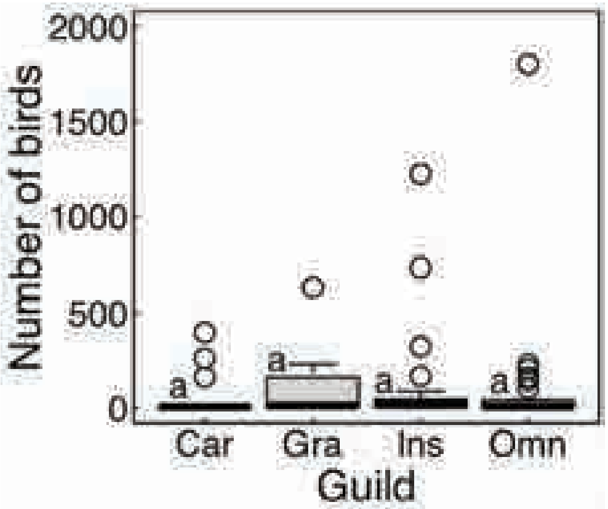

Figure 6. Relative abundance of the birds based on the various feeding guilds. The horizontal black lines in the box indicates the median. The top and bottom edges of each box represent the 75th and 25th percentiles, respectively. The whiskers of the box plot encompass the data within a range of 1.5 times the interquartile range, spanning the upper and lower quartiles. Outliers are indicated by open circles. Identical letters on the box plot signify statistical significance was not found based on non-parametric Kruskal-Wallis test.
Car—Carnivorous | Gra—Granivorous | Ins—Insectivorous | Omn—Omnivorous.

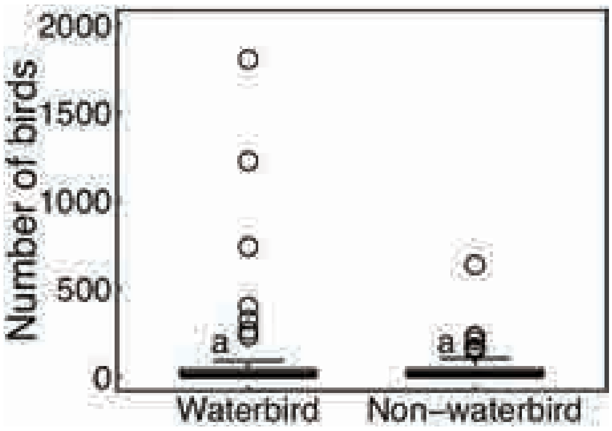

Figure 7. Relative abundance of waterbird and non-waterbird found at the study site. The horizontal black lines in the box indicates the median. The top and bottom edges of each box represent the 75th and 25th percentiles, respectively. The whiskers of the box plot encompass the data within a range of 1.5 times the interquartile range, spanning the upper and lower quartiles. Outliers are indicated by open circles. Identical letters on the box plot signify statistical significance was not found based on non-parametric Mann-Whitney test.

were Ruddy Shelduck (Anatidae), followed by White Wagtail (Motacillidae), River Lapwing (Charadriidae), Oriental Turtle-dove (Columbidae), Plumbeous Redstart (Muscicapidae) and Common Sandpiper (Scolopacidae). The findings imply that the site is relatively rich in avian diversity and richness as evidenced by the detection of birds that belonged to various migration status. Therefore, the Babesa STP holds great potential as a habitat for a diverse population of birds including vagrant, resident and migratory waterbird species.

The family Anatidae, which includes wintering birds such as Ruddy Shelduck, Common Shelduck *Tadorna tadorna*, Common Merganser, Mallard *Anas platyrhynchos*, Red-crested Pochard *Netta rufina*, Eastern Spot-billed Duck *Anas zonorhyncha*, Common Teal *Anas crecca*, Falcated Duck *Mareca falcata*, Northern Pintail *Anas acuta*, Northern Shoveler *Spatulal clypeata*, Gadwall *Mareca Strepera*, Eurasian Wigeon *Mareca penelope*, Ferruginous Duck *Aythya nyroca*, Tufted Duck *Aythya fuligula*, and Garganey *Spatula querquedula*, was found to have the highest RDi value, as previously reported by Tak et al. (2010) and Kumar et al. (2016), which reported a high abundance of the Anatidae family among wetland avifauna communities.

These findings further support the significance of the study site as an important area for avian biodiversity. In the present study, the wintering ducks were mostly seen to inhabit open water and avoided thick vegetation presumably because of limited space and minimal foraging scope (King & Wrubleski 1998; Benoit & Askins 1999).

We observed a large flock of Ruddy Shelduck foraging, resting and roosting at the study site. We also observed Common Merganser foraging in the treatment plant twice. Some conceivable reasons for the substantial number of wintering ducks could be the availability of food resources and size of the wetland (Afdhal et al. 2012; Murray 2014), minimal interference, physical features of wetland habitats (Chatterjee et al. 2020), lack of hunting zones and predators (Kloskowski et al. 2009) at the study site. However, we cannot dismiss the role that the fresh water ecosystem might have played in attracting these birds, especially Ruddy Shelduck, given its close proximity to the STP, or vice versa, as we observed them shuttling between the two during our field visits.

Further, high invertebrate production has also been suggested as one of the key drivers for the occurrence and abundance of waterbirds (Augustin et al. 1999), which could have provided favorable foraging opportunities. Similarly, shorebirds and waders such as Common Sandpiper, Green Sandpiper *Tringa ochropus*, River Lapwing, Grey-headed Lapwing *Vanellus cinereus* and Long-billed Plover *Charadrius placidus* were seen confined to the edges of the STP and on the banks either

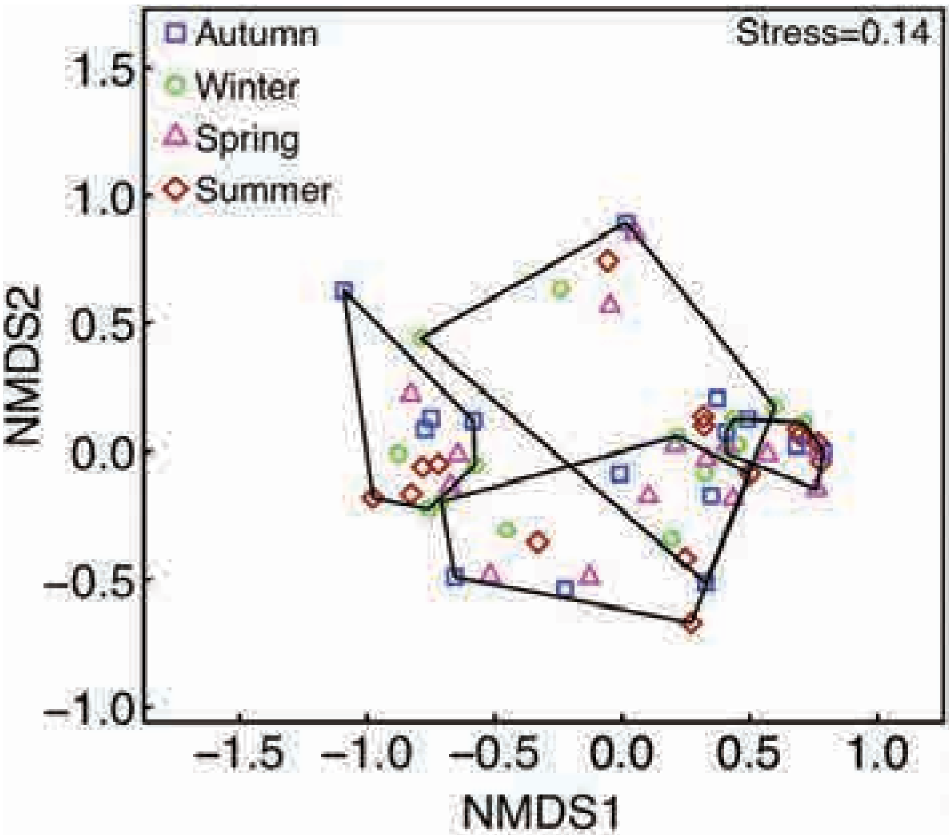

Figure 8. Non-metric multidimensional scaling (NMDS) plot showing dissimilarity in bird species composition across autumn, winter, spring and summer based on the Bray-Curtis dissimilarity matrix of species abundance data with square root transformation. Stress = 0.14.

resting or exploring food resources such as insects, invertebrates, worms and seeds.

The aforementioned findings are in congruence with previous literatures (Muhammad et al. 2018; Luo et al. 2019; Holbech & Cobbinah 2021). Taken together, the results highlights that the Babesa STP is a critical stopover ground and wintering site for many migratory birds which spends as long as six months at the site prior to their summer migration. Perhaps, artificial wetlands have been acknowledged as important migration routes for numerous diving ducks (Kennedy & Mayer 2002). Altogether, that the artificial wetlands hold potential value and can be of importance for migratory waterbird species was reported by Giosa et al. (2018).

Moreover, three 'Near Threatened' waterbird species, namely River Lapwing, Falcated Duck, and Ferruginous Duck, occurred at the study site. The River Lapwing occurred throughout the study period while the Falcated and Ferruginous ducks occurred only during winter (February) and spring (March) months. This indicates that constructed wetlands such as Babesa STP play an indispensable role in conservation and provide important sanctuaries even for threatened species.

Regarding the non-waterbirds, the richness and diversity could be attributed to resources, surrounding habitat and cover along with availability of food (van Biervliet et al. 2020). Indeed, on many occasions we observed non-waterbirds, especially Grey-backed Shrike *Lanius tephronotus* and Common Stonechat *Saxicola maurus*, feed on insects, seeds and fruits, and Eurasian Hoopoe *Upupa epops* forage on edges of the STP as it afforded easy availability of prey.

Likewise, availability of the trees and plants within the vicinity of the study site could have been central to their large assemblages because we observed many of them roost on the branches of the trees and plants. Consistent with this, plant diversity has been shown to exert a positive influence on the bird richness and diversity (Fontana et al. 2011) as it affords microhabitats for roosting, nesting and feeding (Canterbury et al. 1999; Soderstrom & Part 1999).

Interestingly, despite the large avian assemblage there was not any statistically significant difference observed between non-waterbirds and waterbirds, which implies that it might afford a suitable habitat for a large number of avian species. The presence of vegetation for roosting and nesting, open water for foraging and swimming as well as the large occurrence of food resources makes the site attractive for the birds. Taken together, the findings suggest that the study site may function as an important ecological niche for various bird species, including both waterbirds and non-waterbirds.

In contrast, the current study observed statistically significant difference in bird composition between the seasons, in agreement with the findings of Kopij & Paxton (2018). Particularly, the largest differences in bird composition were observed between winter and summer, and between winter and spring. These findings indicate that the dissimilarities in bird compositions across seasons are particularly conspicuous between the dry and monsoon seasons, as well as between the dry and pre-monsoon seasons.

Further, spring and autumn were found to have the highest avian diversity while winter and autumn had the highest species richness compared to spring and summer, respectively. This may be due to seasonal changes in food and resource availability, competition among related species, and predator avoidance strategies (Morin 2011), which may lead to birds utilizing different food sources that vary in quantity and accessibility over time. Additionally, the allocation of resources over time may aid in the coexistence of avian species by allowing for the exploitation of shared resources at different times (Kopij & Paxton 2018). Also, variations in the population and peak abundance of birds across seasons may suggest the migratory patterns of the birds and reveal the direction of migration (Nisbet 1957).

With regard to the feeding guilds, there was no statistically significant difference between the guilds. This statistically insignificant result may be due to the occurrence of a variety of shrubs, flowering trees and diverse array of diets such as fishes, amphibians, reptiles, mammals, and aquatic invertebrates resulting from a large fertility of sewerage treatment plant (Rajpar & Zakaria 2013; Mukhopadhyay & Mazumdar 2019) culminating in the attraction of different guilds. The diversity of feeding guild observed among birds in the vicinity of the study site certainly suggests that it may be an important avian habitat to support various foraging behaviors.

CONCLUSION

Overall, the present study provides a comprehensive assessment of the avian biodiversity present at the Babesa STP. The results reveal that the site harbors a great variety of bird species, including vagrant, resident and migrant birds as well as birds of various feeding guilds. These findings are particularly remarkable given the relatively small size of the study site. Additionally, the findings also underscore the ecological significance of man-made habitats in reinforcing biodiversity, since such ancillary habitats can afford crucial resources and support for a diverse array of species, and act as winter sojourn for migratory birds.

In light of the findings of this study, it is recommended that concerned authorities and policymakers take further action to safeguard the site as it is important for bird conservation. For instance, a valuable intervention measure for the area may be fencing to keep away potential predators such as stray dogs, which are quite common in the area. Additionally, certain points may be identified as photography spots to minimize human-induced disturbance to the birds. Otherwise, apart from serving as a suitable area for recreation, bird watching and scientific study, the site can also be a great source of educational opportunities for students, teachers, and the general public interested in learning about the features and importance of constructed wetlands in sustaining wildlife habitats and biodiversity (Semeraro et al. 2015). Further research is warranted, especially concerning the underlying factors that trigger large assemblages of birds at the site.

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Threatened Taxa

Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23487–23498

ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)

https://doi.org/10.11609/jott.8416.15.7.23487-23498

#8416 | Received 20 February 2023 | Final received 30 May 2023 | Finally accepted 23 June 2023

COMMUNICATION

Proximate nutrients of selected forage and the diet composition of adult elephants in Udawalawe National Park, Sri Lanka, a preliminary study

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Abstract: Asian Elephants feed predominantly on grass. The comparative nutritional contribution of grasses and other elephant forage is not known. Therefore, the proximate nutrition of food plants selected by elephants, and the relationship of their diet composition to body condition and gender were examined in this study. Proximate analysis was conducted on 11 plant species recognised upon 66h of opportunistic focal animal sampling. Five species among them were grasses, including the invasive *Megathyrsus maximus*. The micro-histological composition of freshly collected dung from 26 identified elephants was assessed against their body condition and gender. Associations, comparisons, and hypotheses were tested. Dicots were significantly high in dry matter and low in moisture, while monocots were high in moisture and low in dry matter ($p < 0.001$). The average monocot: dicot ratio was 1: 0.73 in elephant diet. However, it was observed that the monocot composition in the male diet was significantly higher than dicots ($p < 0.001$), while there was no significant difference in the female diet composition. Elephant body condition did not show any correlation with the abundance of monocot or dicot plant tissues. The preliminary study implies that dry matter nutrients in dicots and moisture in monocots influence diet selection of elephants. Their diet composition was associated with gender but did not correlate with body condition. *M. maximus* was not outstanding in nutrition from the selected plant species.

Keywords: Asian Elephant, body condition, *Elephas maximus maximus*, food selection, gender, mammals, nutrition.

Abbreviations: UNPSL—Udawalawe National Park of Sri Lanka | DM—Dry matter | BCS—Body condition score.

Editor: Heidi Riddle, Riddle's Elephant and Wildlife Sanctuary, Arkansas, USA. **Date of publication:** 26 July 2023 (online & print)

Citation: Hemachandra, I.V.D., C.D. Wijayarathna & P.N. Dayawansa (2023). Proximate nutrients of selected forage and the diet composition of adult elephants in Udawalawe National Park, Sri Lanka, a preliminary study. *Journal of Threatened Taxa* 15(7): 23487–23498. https://doi.org/10.11609/jott.8416.15.7.23487-23498

Funding: This research was partially funded by the University of Colombo.

Competing interests: The authors declare no competing interests.

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Author contributions: All authors contributed to the study conception and design. Material preparation, data collection and analysis were performed by I.V.D. Hemachandra, C.D. Wijayarathna, and P.N. Dayawansa. The first draft of the manuscript was written by I.V.D. Hemachandra and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

Research permission: Permission was obtained for this research from the Department of Wildlife Conservation, Sri Lanka for observation of elephants, collection of elephant dung and plant samples under the Permit No: WL/3/2/55/19.

Acknowledgement: Authors acknowledge Mr. M.D.S. Salgado and Mr. Dilup Chandranimal Perera for their expert assistance in identifying plant species, and Emeritus Prof. W.D. Rathnasooriya for reviewing the manuscript and providing valuable comments for improvement. Authors would also like to acknowledge the Department of Wildlife Conservation for granting permission to conduct this research in the Udawalawe National Park, the herbarium of the Plant Science Department of University of Colombo for allowing to refer specimens, and the University of Colombo for partially funding the research.

INTRODUCTION

Elephants are bulk feeders with an ability to selectively feed on different forage using their highly specialised trunk (McKay 1973; Eisenberg 1980; Owen-Smith 1988; Dumonceaux 2006). They are generalised mixed feeders (Shoshani & Eisenberg 1982; Fernando & Leimgruber 2011). These monogastric megaherbivores are colonic hindgut fermenters with a very short food retention time due to a relatively short gut (Greene et al. 2019). Studies conducted on the diet of Asian Elephants in the wild include identification of forage plants, their availability and foraging nature, and the study of foraging behaviour (Eisenberg 1980; Steinheim et al. 2004; Chen et al. 2006; Pradhan et al. 2008; Baskaran et al. 2010). Few studies have been carried out on nutrition of their natural diet (Das et al. 2014; Lihong et al. 2007; Borah & Deka 2008; Santra et al. 2008; Koirala et al. 2018). Asian Elephants are observed to prefer and feed more on grasses (Samansiri & Weerakoon 2007; Fernando & Leimgruber 2011; Alahakoon et al. 2017).

It is reported that Sri Lankan elephants spend about 75% of their daily activity budget on feeding, while an adult elephant feeds on about 150 kg and defecates about 80 kg of forage per day (Vancuylenberg 1977; Eisenberg 1980). Feeding behaviour and foraging ecology of elephants, including plant identification and their availability, have also been conducted in Sri Lanka (McKay 1973; Vancuylenberg 1977; Samansiri & Weerakoon 2007; Angammana et al. 2015; Alahakoon et al. 2017). The Sri Lankan Elephant's large diet breadth has been examined. A total number of 116 species of food plants of elephants belonging to 25 families were recorded from northwestern Sri Lanka by Samansiri & Weerakoon (2007), while a diet breadth of 63 food plants was identified by Alahakoon et al. (2017) from Udawalawe National Park of Sri Lanka (UNPSL). Despite, there is a lacuna in the study of nutrition of the natural diet of Sri Lankan elephants.

It has been opined that recently reported observations of elephants with poor body conditions in UNPSL could be due to rapid reduction of the distribution of Guinea Grass (*Megathyrsus maximus*) (Anver 2015; Fernando 2015b; Wijesinghe 2016). *Megathyrsus maximus* is an invasive species introduced as fodder for livestock (Panwar & Wickramasinghe 1997; Wisumperuma 2007). Hence it is important to understand whether the reduced extent of Guinea Grass could affect elephant body condition. Accordingly, this study was conducted with the following primary objectives: (a) Studying the proximate nutrients of selected plant materials in the diet of elephants at UNPSL; (b) Understanding the diet composition in relation to gender and body condition of elephants at UNPSL; and (c) Obtaining an ecological insight into the relationship between diet composition of elephants and the nutritional composition of their feeding materials. Also, the secondary objective of this study was to compare the nutritional value of invasive *M. maximus* with the selected food plants, especially the other grass species.

MATERIALS AND METHODS

Study site

Udawalawe National Park of Sri Lanka (UNPSL) has an extent of 308.2 km^2. It is located between 6.4167^{0}N & 6.5833^{0}N, 80.7500^{0}E & 81.0000^{0}E in the intermediate zone between wet zone and dry zone (Figure 1). The location experiences dry periods between a narrow rainy period (February to April) and a longer rainy season from end of August to December. The mean annual rainfall of UNPSL is about 1,524 mm (Angammana et al. 2015) and Udawalawe and Mau Ara reservoirs are found within it. Major vegetation types of UNPSL are comprised of intermediate zone to dry zone transitional monsoon moist forests in the northern part, dispersed grasslands, scrubs, and different stages of succession (Panwar & Wickramasinghe 1997; Alahakoon et al. 2017).

UNPSL is the third most visited national park of Sri Lanka (Kariyawasam & Sooriyagoda 2017). It is well known for easy sighting of elephants and has been recorded to host 800–1,160 elephants (de Silva et al. 2011).

Permission was obtained from the Department of Wildlife Conservation, Sri Lanka, for observation of elephants, collection of elephant dung and plant samples (Permit No: WL/3/2/55/19).

Determination of nutritional composition in forage

Sample collection

Upon conducting opportunistic focal animal sampling for 66 hours in August 2019, 11 plant species were selected based on the observed foraging behaviour of Sri Lankan elephants *Elephas maximus maximus* inhabiting the site. Selective feeding of mammalian herbivores extends further from plant species to specific plant parts (Owen-Smith & Chafota 2012). Therefore, plant parts varying from complete aerial body, stem, leaves, to fruits, were collected according to the choice of plant varieties by the elephants. Plant parts were selected considering the acceptance of the plant from an observed site, based on the elephant's behaviour, as described in Owen-Smith & Cooper (1987).

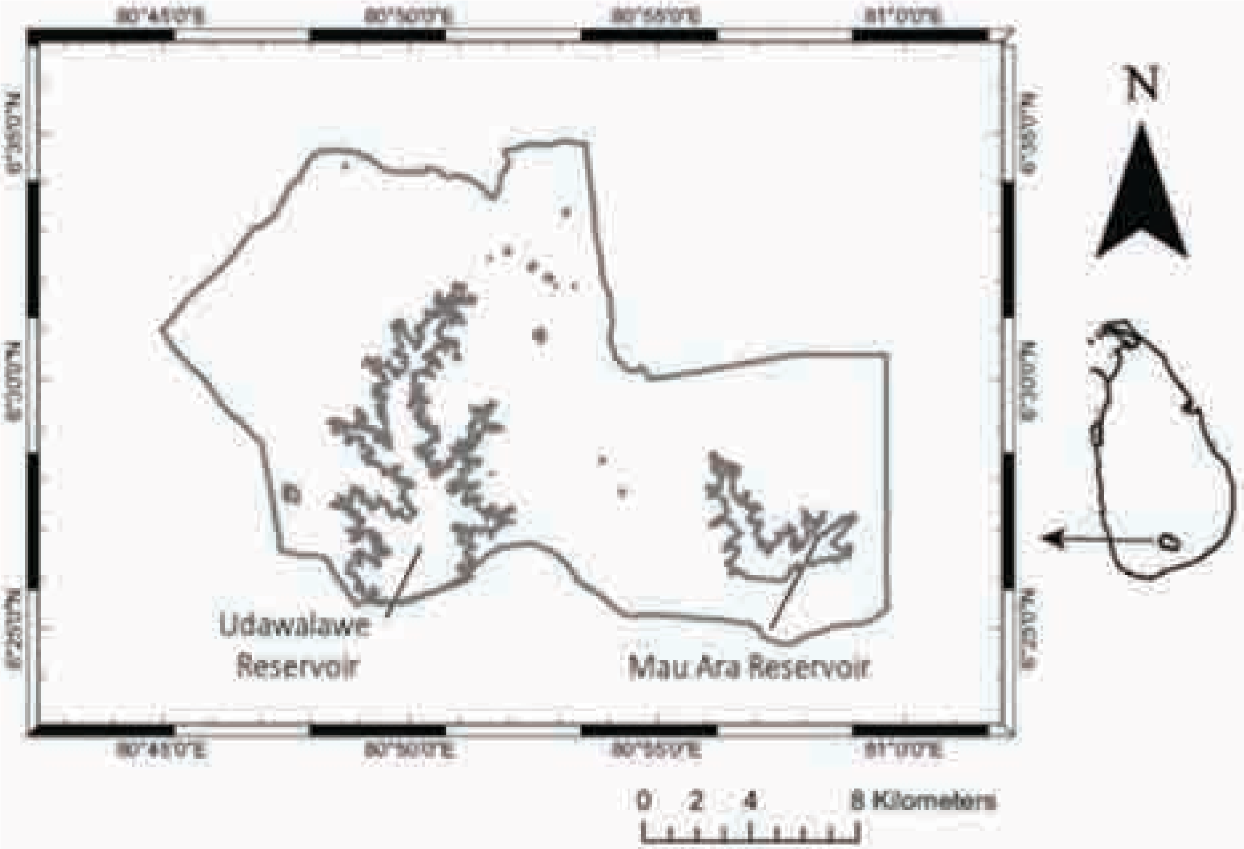

Figure 1. Udawalawe National Park of Sri Lanka (mapped by authors).

The acceptance value was calculated by dividing the utilised number of plants from the available number of plants of a species from the observation site (Owen-Smith & Cooper 1987). Browsed species were counted as individual plants, adapting the method to count grazed species as patches (1x1 m^2) due to their numerous availability and maximum utilisation of their aerial body. It was assumed that the patches of small herbs and grasses were not heavily mixed and represented the nearest randomly missed out/ dropped plants during feeding. The extent of the observation site was determined according to the utilisation area of the focal elephant until it moved out of sight. Plants that had an acceptance rate above 0.5 were selected for sample collection.

Most of the plant species were identified in situ, however, when it was difficult to identify, herbarium samples of the unidentified species were obtained for identification using guides, reference herbarium collections, and through expert assistance. About 200 g of fresh plant matter was collected into re-sealable plastic bags.

The amount of nutrients in plants can differ among habitats, seasons, and maturity of the plant (Rothman et al. 2012; Das et al. 2014; Koirala et al. 2018). Hence the plant parts were selected from the same plants that the elephants were feeding from. For grasses and herbs, samples were collected from the same site as the same plant could not be obtained due to total consumption by the elephants.

Sample preparation

The nature of the consumed plant part, such as maturity, and the exact way in which the plant part was processed by the elephant was also considered during sample preparation (Dierenfeld 2006; Rothman et al. 2012; Ranjeewa et al. 2018). For example, it was observed in the field that elephants feed on thorny *Limonia acidissima* stems only after removing thorns with the aid of their trunks before ingestion. Mature *Bauhinia* legumes were analysed, and the complete legume was used without separating seeds during laboratory analysis. It was presumed that the entire legume was processed in the gut as manual dissection of dung analysis did not reveal any traces of the legume. The digestion of the legumes in elephants is not known, although *Bauhinia* seeds have been found in elephant dung (Chathuranga &

Ranawana 2017).

Collected samples were washed and allowed to dry in the shade before being used in analysis of nutrients. Long twigs and stems were cut to small parts. Prepared plant materials were mixed well before obtaining a subsample for nutritional analysis, to ensure random sampling.

Sample analysis

The amount of moisture, dry matter (DM), ash content, crude protein and crude fats was measured in the plant samples collected from the selected species and quantified amounts were expressed as a percentage of initial mass (w/w). It was assumed that the remaining mass amounts for the total carbohydrates in the sample and it was estimated by substituting the amount of other measured nutrients for the following modified equation adopted from Maclean et al. (2003).

Total carbohydrate % = 100% – ([crude protein + crude fats + water + ash content] %)

All analysed nutrient masses were weighed using an analytical balance BSA223S-CW (max 220 g, least count = 1 mg). The results of analysis were expressed as fed (wet) and dry matter percentages. Analyses were triplicated.

Dry matter/ moisture and ash content

Subsamples of 10 g were measured from each of the collected plant samples and then dried in an air circulating oven at 70–80°C until a constant mass of dry biomass was obtained (Levett et al. 1985). Moisture content was calculated by deducting the dry biomass from the wet biomass.

Oven dried samples were transferred to porcelain crucibles, dried at 550° C for 4 h in a muffle furnace (Model HD-230, Spain) (Richards 1993). The mass of the obtained ash was weighed, to express the percentage wet mass.

Proteins

Proteins were extracted from the samples of 0.5 g of plant material using the salt/ alkaline extraction method with modifications. The prepared plant protein samples were analysed by mixing 1 ml of plant extract with 4.5 ml of Biuret reagent against the blank sample using an UV–Vis spectrophotometer at 545 nm wavelength. The obtained absorbance values were traced to determine the respective concentrations of protein in the samples, using a standard curve obtained for known concentrations of Bovine Serum Albumin (BSA) with Biuret reagent within the range of absorbance (545 nm) at 0.2–0.7.

Crude fats

Fresh samples of 5 g were randomly picked from the collected plant samples. Solvent extraction (AAFCO Lab Methods and Services Committee 2014) with diethyl ether was performed for the plant samples.

Micro-histological composition of dung

Dung samples were freshly collected soon after defecation from 26 elephants, out of a total of 509 individual elephants assessed in UNPSL from August to November 2019. The sampling period covered both wet and dry seasons. Two boluses of dung from each elephant's dung pile were collected in a re-sealable plastic bag within a short period upon defecation as soon as the elephants left the study site. Gender and age of the elephants were determined according to Varma et al. (2012). The body condition scoring (BCS) method used in this study replicated the modified Wemmer et al. (2006) method used by Ranjeewa et al. (2018) previously in UNPSL. The visual body condition scoring method which assesses fat deposition in seven prominent areas of the elephant's body considered the appearance of the following body areas: temporal depression at the head, distinction of shoulder blades at the scapular area, prominence of ribs at the thoracic area, the area immediately in front of the pelvic girdle at the flank, the spine between shoulder and pelvic girdle at the thoracic spine, the spine between the pelvic girdle and base of tail at the lumbar spine, and the pelvic girdle at the pelvic area. The recorded body condition scores were normally distributed from a minimum of three (3) to a maximum fourteen (14) within the range of the methodology (0–14). The elephants were identified individually by the morphological features on their body (depigmentation, lumps, wounds, ear tears, ear shape, tail characters, etc.) as described in Fernando et al. (2011) and Vidya et al. (2014).

The ratio of the monocotyledonous and dicotyledonous tissues of dung samples was determined microscopically. A subsample of 20 g of dung was obtained and processed according to Fernando et al. (2016) for the microscopic analysis of plant tissues in elephant dung. A scraping of the final residue was observed under the light microscope at x100 magnification, and the monocotyledonous and dicotyledonous tissues were counted using a Sedgewick rafter counting chamber. Each subsample was observed in triplicates to determine an average count of monocotyledonous and dicotyledonous tissues.

Statistical analysis

To test the hypotheses, the dung analysis and nutrition analysis data were checked for normality and statistically tested using IBM SPSS Statistics version 26 software. The relationship of the visual body condition score and the gender of wild elephants (n = 26), with the monocotyledonous and dicotyledonous tissue count in their dung samples was analysed with Pearson correlation test and chi-square test for association, respectively. The sample means between the monocotyledonous and dicotyledonous tissue counts in each gender group, as well as the sample means of tissue counts of each plant group between the genders was compared by two sample t tests to further understand the relationship between the diet composition and the gender of elephants.

In the nutritional analysis of selected food plants, the mean values and standard errors were calculated for each analysed plant species as well as the plant group (monocotyledonous and dicotyledonous). The composition of moisture, dry matter in the monocotyledonous and dicotyledonous plants was compared by Mann-Whitney test. The 'as fed' and 'dry matter' compositions of each proximate nutrient (ash content, crude protein, crude fats, and total carbohydrates) between the two groups of monocotyledonous and dicotyledonous plant samples were also compared using Mann-Whitney test or two sample t tests according to the normality of data distribution.

To examine whether *Megathyrsus maximus* had a significantly different nutritional contribution from other selected grasses, the nutrition composition of grasses was compared using the Kruskal-Wallis test and post hoc pairwise comparison.

RESULTS

Plant sample collection

Five monocotyledonous plants which were all grasses (Family Poaceae) and six key dicotyledonous plants were selected for the nutritional analysis based on observation of elephant foraging behaviour and are shown in Table 1.

Forage nutrition

The nutritional composition of analysed plant materials was expressed in mass percentages in both wet basis and dry basis (DM) as given in Table 2. Figure 2 presents the moisture content, total dry matter, and other nutrients (ash content, crude proteins, crude fats, total carbohydrates) in wet basis, while Figure 3 presents the dry basis of the nutrients in the studied plant samples.

It was observed that monocotyledonous plants (Mean±SE: 74.76±0.96) had a significantly higher amount of moisture over dicotyledonous plant parts (42.4±3.30) consumed by elephants. DM in dicotyledonous plants was significantly higher compared to monocotyledonous plants (P <0.001). The as fed composition of ash content (7.80±1.40) and total carbohydrates (29.50±4.00) in the dicotyledonous plants was significantly higher than the as fed ash content (3.10±0.20) and total carbohydrates (14.17±0.90) in monocotyledonous plants (P <0.001). There were no significant differences in the dry matter compositions of nutrition between monocotyledonous and dicotyledonous samples.

Megathyrsus maximus was similar to several other grasses assessed in this study for each proximate nutrient either in as fed or dry matter composition.

Table 1. Selected plants and different parts used for the analysis.

Group	Plant (Scientific name and Common name)	Analysed part	Foraging method by elephant	Acceptance value
Monocotyledonous	*Megathyrsus maximus* (Guinea Grass)	Total aerial body	Grazed grass	0.67
	Lepturus radicans	Total aerial body	Grazed grass	0.79
	Cyrtococcum spp.	Total aerial body	Grazed grass	0.88
	Bouteloua dactyloides (Buffalo grass)	Total aerial body	Grazed grass	0.72
	Garnotia fergusoni	Total aerial body	Grazed grass	0.71
Dicotyledonous	*Phyllanthus polyphyllus*	Leaves	Grazed shrub	0.85
	Achyranthes aspera (Devil's horsewhip)	Total aerial body	Grazed herb	0.67
	Cryptolepis buchananii	Leaves from a young climber	Browsed climber	0.73
	Bauhinia racemosa	Mature dried fruit (legume)	Browsed/ Picked from ground	0.62
	Ziziphus oenoplia (Jackal Jujube)	Leaves from young tree	Browsed shrub	0.58
	Limonia acidissima (Woodapple)	Leaves and stem from young tree	Browsed tree	0.55

Table 2. Mass percentage of nutritional composition of analysed plant samples (sample size: 3).

Group	Plant sample	Percentage (%) (Mean±SE)									
		Moisture content	Dry matter (DM)	Ash content		Crude protein		Crude fats		Total carbohydrates	
				As fed	DM	As fed	DM	As fed	DM	As fed	DM
Monocotyledonous	*Megathyrsus maximus*	73.90 ±1.21	26.10 ±1.21	3.36 ±0.70	13.20 ±3.30	5.84 ±1.69	22.01 ±6.02	0.35 ±0.04	1.31 ±0.11	16.55 ±1.29	63.48 ±4.49
	Lepturus radicans	75.87 ±0.91	24.13 ±0.91	3.50 ±0.15	14.5 ±0.12	4.87 ±0.67	20.04 ±1.98	0.35 ±0.02	1.44 ±0.06	15.41 ±0.17	64.02 ±2.00
	Cyrtococcum sp.	79.51 ±0.04	20.49 ±0.04	3.64 ±0.21	17.76 ±0.98	5.56 ±0.66	27.11 ±3.19	2.83 ±0.15	13.79 ±0.71	8.46 ±0.97	41.34 ±4.82
	Bouteloua dactyloides	75.57 ±1.19	24.43 ±1.19	2.75 ±0.41	11.16 ±1.29	8.31 ±0.85	33.86 ±2.36	0.05 ±0.00	0.20 ±0.02	13.32 ±0.48	54.78 ±3.62
	Garnotia fergusoni	68.96 ±0.39	31.04 ±0.39	2.25 ±0.18	7.24 ±0.61	11.06 ±0.60	35.7 ±2.38	0.61 ±0.06	1.98 ±0.22	17.11 ±1.03	55.08 ±2.64
Dicotyledonous	*Phyllanthus polyphyllus*	60.07 ±0.56	39.93 ±0.56	2.97 ±0.29	7.45 ±0.79	25.33 ±0.37	63.44 ±0.63	1.64 ±0.40	4.08 ±0.93	9.99 ±0.03	25.02 ±0.27
	Achyranthes aspera	48.20 ±0.05	51.80 ±0.05	7.84 ±0.14	15.13 ±0.28	3.45 ±0.40	6.67 ±0.78	23.72 ±0.68	45.78 ±1.27	16.79 ±0.99	32.42 ±1.95
	Cryptolepis buchananii	61.23 ±0.08	38.77 ±0.08	2.89 ±0.13	7.45 ±0.35	22.51 ±1.72	58.05 ±4.35	0.85 ±0.09	2.18 ±0.23	12.52 ±1.52	32.31 ±3.97
	Bauhinia racemosa mature legume	19.40 ±0.49	80.60 ±0.49	5.43 ±0.79	6.74 ±1.16	45.75 ±3.58	56.79 ±2.60	0.29 ±0.03	0.36 ±0.22	29.13 ±3.30	36.11 ±3.87
	Ziziphus oenoplia leaves	38.00 ±0.31	62.00 ±0.31	20.47 ±2.83	32.97 ±4.42	5.46 ±1.04	8.80 ±1.66	0.30 ±0.04	0.49 ±0.06	35.77 ±3.06	57.74 ±5.25
	Limonia acidissima leaves	34.27 ±0.82	65.73 ±0.82	9.10 ±0.62	13.82 ±0.78	11.39 ±1.61	17.39 ±2.66	3.54 ±0.11	5.39 ±0.17	41.71 ±1.80	63.41 ±1.94
	Limonia acidissima stem	27.79 ±0.73	73.21 ±0.73	5.14 ±0.17	7.12 ±0.28	6.26 ±0.81	8.70 ±1.21	0.22 ±0.01	0.31 ±0.02	60.59 ±1.61	83.88 ±1.47

Micro-histological analysis of elephant dung

Among the 26 individual elephants, 10 were males and 16 females, and 24 were adult elephants while two were subadult males. The average ratio of monocotyledonous (grasses): dicotyledonous tissues in dung was 1: 0.73 (57.95: 42.04±3.78 %) in average. The relative abundance of monocotyledonous tissues (0.58±0.03) was significantly higher than that of dicotyledonous tissues (0.42±0.03) ($p < 0.001$) in the examined dung samples. There was no significant difference between the abundance of monocots ($p = 0.877$) or dicots ($p = 0.815$) between the wet and dry seasons.

There was an association between the gender of the elephants and the type of tissues (monocotyledonous, dicotyledonous) found in their dung ($p = 0.041$, Pearson chi square = 4.196). The relative abundance of monocotyledonous tissues (64±4.8%) was significantly higher than dicotyledonous tissues (36±5.0%) in dung samples obtained from males ($P < 0.001$). However, based on the dung analysis, there was no significant difference between the abundance of monocotyledonous and dicotyledonous tissues detected in dung samples of female elephants.

There was no significant difference ($p = 0.065$) between the relative abundance of monocotyledonous tissues detected in the dung samples of male and female elephants. Also, a significant difference was not observed ($p = 0.132$) between the relative abundance of dicotyledonous tissues detected in the dung samples of male and female elephants.

The average body condition of the focal elephants was 8.15±1.73. The lowest BCS recorded was three (3) while the highest was fourteen (14). The body condition score of the elephants had no significant correlation with the abundance of monocotyledonous tissues or the abundance of dicotyledonous tissues. Neither did the relative abundance of monocotyledonous or dicotyledonous tissues correlate with the body condition score of the elephants. This result was consistent when each gender group (male and female) was considered separately. There was no correlation between the body condition and the abundance of monocots or dicots within either gender group.

DISCUSSION

This study is the first comparative analysis of nutrition between the grasses and other forage of wild elephants in Sri Lanka. Although many studies have reported the

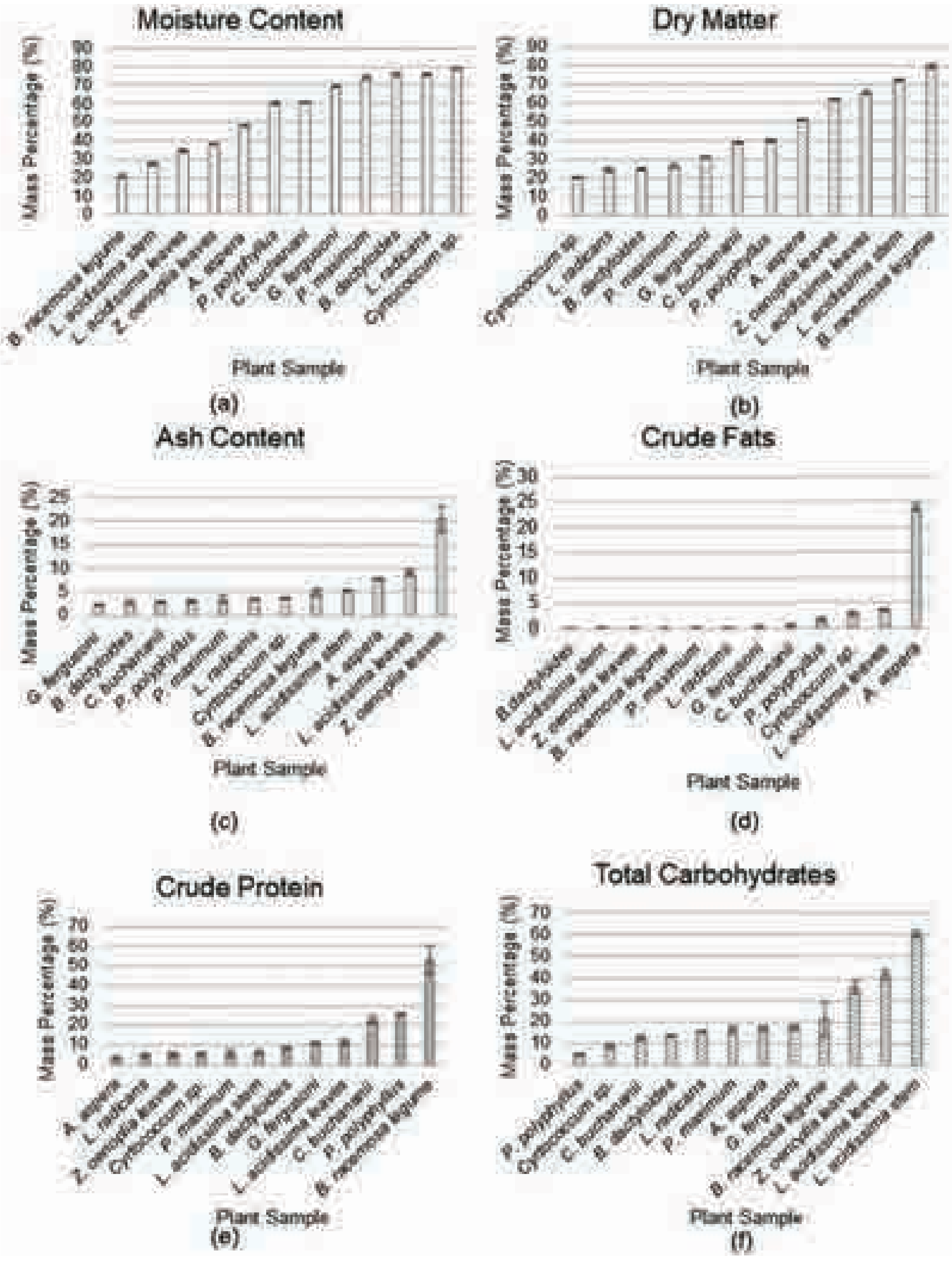

Figure 2. Percentage (w/w) (%) in as fed basis: a—Moisture content | b—Dry matter | c—Ash content | d—Crude fats | e—Crude protein | f—Total carbohydrates.

ratio of monocotyledonous to dicotyledonous tissues in elephant dung (Steinheim et al. 2005; Samansiri & Weerakoon 2007; Koirala et al. 2016), this is also the first study to report dung composition of identified adult wild elephants from Sri Lanka, enabling the comparison of their body condition and gender with their diet composition revealing important novel findings. According to the dung analysis results, the diet preference of elephants in UNPSL is dominated by monocotyledonous plants, represented mainly by grasses. However, the results suggest a difference in the diet composition of the males and females. There was no relationship between the body condition of elephants and the plant type. The proximate analysis revealed that dicotyledonous food plants are more nutritious than monocotyledonous grasses as expected. But the moisture content of grasses was unexpectedly high, suggesting that the preference for grasses may be influenced by the feed moisture as well. *Megathyrsus maximus* was similar to other selected grass species in nutrition. Altogether, these results suggest that the disappearance of invasive *Megathyrsus maximus* from UNPSL could not affect the body condition of the elephants.

Proportions of Monocot and Dicot Tissues

The results are consistent with previous research that suggests that the Asian Elephant is adapted to a natural diet high in grass. Samansiri & Weerakoon (2007) had also reported that monocotyledonous tissues were dominant in the dung collected from elephants in northwestern areas of Sri Lanka. Alahakoon et al. (2017) observed that elephants in UNPSL show a higher behavioural frequency in feeding grasses. The same has been observed in Assam, India (Borah & Deka 2008). Grasses are accessible to elephants of all age groups (Baskaran et al. 2010). Juveniles predominantly forage on grasses (Samansiri & Weerakoon 2007). The diet composition of elephants has been observed to change among seasons in other countries (Steinheim et al. 2005; Chen et al. 2006; Lihong et al. 2007; Pradhan et al. 2008; Baskaran et al. 2010; Koirala et al. 2018). Generally, the Asian Elephant foraging is considered to be dominated by grazing during the wet season and browsing during the dry season (Sukumar 1990; Baskaran 2010). In Nepal, it has been observed that while browsing is dominant during dry season, both browsing and grazing are equally important during the wet season (Koirala 2016). However, in Sri Lanka, especially UNPSL, it has been reported that grasses have remained dominant in the diet constantly as they regenerate during each season, as usual during wet season and as a special occurrence on exposed tank beds of the main two reservoirs within UNPSL during the dry season (Alahakoon et al. 2017; Ranjeewa et al. 2018; Sampson et al. 2018). Hence, the absence of a significant difference in monocots or dicots between the wet and dry seasons is possibly due to the influence of climatic factors and geographic features at UNPSL.

The dung composition and the gender biased access to resources

No reported information was found on the diet composition and gender of elephants in literature and an interesting difference between the genders was observed in the present study. Adult male and female elephants indicate distinct gender roles in the wild. Generally, female elephants live in family units while adult male elephants are solitary animals (McKay 1973; Schulte 2006). The same social arrangement was observed in the UNPSL during this study. Sri Lankan elephants avoid competition for food (Yapa & Rathnavira 2013). McKay (1973) reported that Sri Lankan elephant herds stay separated from other herds in the same area and the female movement rates are significantly slower when moving, while feeding, owing to needs to nurture and care for the young. Accordingly, the amounts and flexibility of food choice available for female elephants in herds are limited in comparison to solitary males. Male elephants are also accused of raiding crops which mainly involve monocotyledonous plants such as paddy *Oryza sativa*, maize *Zea mays* of family Poaceae, and palms (Arecaceae) such as coconut *Cocos nucifera* and kitul *Caryota urens* that are generally found associated with human settlements (Samansiri & Weerakoon 2007; Fernando 2015a).

The nutritional needs of animals change with their stage of life. The young and juvenile need nutrition for weight gain, bone and muscle development, while lactating and expectant animals require additional nutrition for nourishing the young (Birnie-Gauvin et al. 2017; Bechert et al. 2019). In Argali *Ovis ammon*, males have been identified to select abundant forage of lower quality (grasses and forbs) and females to select higher quality forage (forbs and shrubs) to achieve energy requirements for nursing and gestation (Li et al. 2018). Consuming more and different types of food plants that are high in nutritional quality minimizes the animal's effort for finding nutritious food (Owen-Smith 1988; Shannon et al. 2006). Moisture also assists digestion and lactation of females to nurse calves (Beede 2005; Van Weyenberg 2006). Accordingly, it could be inferred from the results that both monocotyledonous grasses and diverse dicotyledonous plants are equally important in the diet composition of an adult female elephant due to their behavioural role. Therefore, the difference in dung composition results in males and females is suggested to be due to behavioural differences affecting food selection of the two genders.

Nutritional composition

The dicotyledonous plants were significantly higher in dry matter nutrition than the monocotyledonous grasses, although the diet composition of the Asian elephants is dominated by monocotyledonous plants. This finding is consistent with previous reported studies

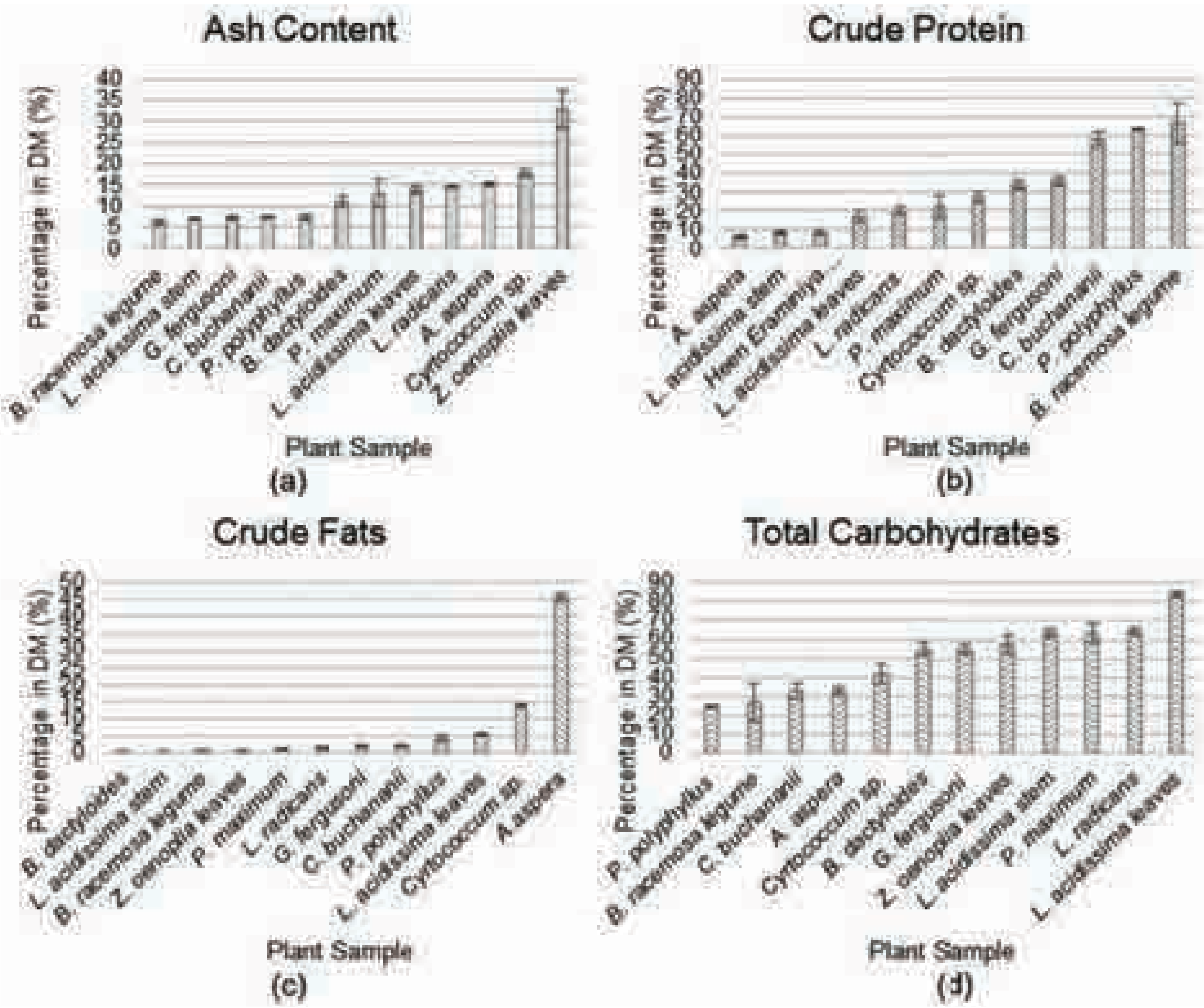

Figure 3. Percentages (w/w) (%) in dry matter basis: a—Ash content | b—Crude protein | c— Crude fats | d—Total carbohydrates.

on elephant nutrition with dicotyledonous plants occupying the highest values for various nutrients (Chen et al. 2006; Lihong et al. 2007; Das et al. 2008; Santra et al. 2008; Borah & Deka 2014). In contrast, the grasses indicated an unanticipated significantly high moisture content (about 70% w/w).

Previous studies conducted on the nutrition of elephant forage have focused on dry matter as that accounts for providing energy to the animal (Chen et al. 2006; Borah & Deka 2007; Lihong et al. 2007; Santra et al. 2008; Rothman et al. 2012; Das et al. 2014; Koirala et al. 2018). Although Santra et al. (2008) present moisture composition, the selected plant parts are limited to browsed plant parts identified from signs of plant damage. This is the first report on the moisture content of both grazed and browsed plant species of elephants.

Feeding large quantities of grass of low nutritional quality and their rapid passing through the gut by large herbivores is recognised as a mechanism of gaining more energy from low quality feed abundant in the environment (Bell 1971; Owen-Smith 1988; McArthur 2014). However, elephants are known to select food from their environment despite their availability (Koirala et al. 2016; Birnie-Gauvin et al. 2017). Therefore, the high moisture in the grass could be an additional incentive for the Sri Lankan elephant that mostly inhabits the dry zone, to select more grasses from their environment. Moisture contributes to the palatability of forage which is a factor in selection and rejection by elephants (Lihong et al. 2007; Santra et al. 2008; Das et al. 2014). Elephants have a high utility rate of water with limited ability to concentrate urine and water loss occurring from frequent urination and defecation (Ratnasooriya et al. 1994; Cheeke & Dierenfeld 2010). Freshly defecated elephant dung has been reported to hold 45–75% (w/w) water content (McKay 1973). The amount of moisture and water holding capacity in feed intake assists digestibility, passage of materials through the gut, and defecation as well (Van Weyenberg et al. 2006). African Elephants have been reported to increase woody parts in their diet during the dry season as the stem and pith of woody plants contain more water content (Owen-Smith 1988; Rothman et al. 2012; Greene et al. 2019). Horses are considered to be closest to elephants in the digestion physiology (Bechert et al. 2019; Greene et al. 2019). Captive horses have also been reported to select hay samples with more moisture and hay wetting behaviour (Müller & Udén 2007; Muhonen et al. 2009; Harris et al. 2016; Müller 2018). Hence, the high moisture content in grass influences preference and selection by elephants.

As elephants are hindgut fermenters, it is considered that they are benefitted from more fermentable feed due to limited digestion of fibre in their gut. The fibre in grass could draw water which is important for the fermentation process required for digestion in the hindgut (Sneddon & Argenzio 1998; Muhonen et al. 2009; Bechert et al. 2019).

Body Condition Score

The relationship of the elephant body condition with their diet composition has not been described previously. The results of this study do not support previous inferences that the availability of grass in the environment supports better body condition of elephants (Ranjeewa et al. 2018). According to Ranjeewa et al. (2018) the average body condition scores of elephants are higher during the dry seasons as more grass grows on the exposed tank bed due to receding water levels. However, according to this study, the relative abundance of monocotyledonous tissues (grasses) in their diet does not correlate with their body condition. Hence the availability of more grasses, especially a single grass species such as *Megathyrsus maximus* in the environment could not be considered as a contributing factor to the elephant body condition.

***Megathyrsus maximus* at UNPSL**

Megathyrsus maximus was not outstanding in nutrition from the other selected plants. Pairwise comparison between the five selected grass species revealed that *Megathyrsus maximus* was nutritionally similar to one or few of the other four grasses (*Bouteloua dactyloides, Cyrtococcum* sp., *Garnotia fergusoni*, *Lepturus radicans*) for the different proximate nutrients analysed, both in as fed and dry matter basis. A study conducted from December 2005 to January 2007 states that 67% of elephant sightings and feeding behaviour (28.9%) observations at UNPSL were made in *Megathyrsus maximus* grasslands that had occupied 39% of the land area of UNPSL (Alahakoon et al. 2017) unlike today where it is limited to a small patch of 0.13 km^2 near the entrance (less than 1% of the area). *Megathyrsus maximus* is a tall grass while other studied grasses were short. Its large size and biomass compared to other smaller ground hugging grasses is the reason for elephants' preference and choice (Fernando 2015b). Elephants are generalists with a large diet breadth. They are bulk feeders and do not linger at one plant species but move ahead through available choices giving it more access to choose food from the environment (McKay 1973). It is reported that they spend more time feeding on short grasses than long grasses (McKay 1973). It had been observed that elephants avoid areas of high *M. maximus* abundance while indicating a positive correlation with short grasses (Sampson et al. 2018). Thus, it could be presumed that Guinea grass does not have an effective nutritional influence for elephant diet in UNPSL.

The dung analysis did not identify *M. maximus* separately, even though the monocotyledonous and dicotyledonous tissues could be distinguished. Presuming that the monocotyledonous tissues in elephant diet are mainly represented by grass according to the vegetation in the UNPSL (DWC 2008), as there was no linear relationship between the abundance of either tissue type with body condition, although there was a significantly high abundance of monocots, it could be concluded that the amount of grass in the diet has no effect on body condition of elephants. Hence, the findings of this study challenge the notion that the reduced distribution of invasive Guinea Grass (*M. maximus*) was the reason for poor body condition of elephants at UNPSL.

Information on dietary choice and differences in elephants are essential for informed decision making in their conservation and management. The elephants in UNPSL preferred grasses, but demonstrated a difference in the food plant selection between the genders which could be attributed to their gender biased behaviour. As generalist megaherbivores with a large diet breadth (Fernando & Leimgruber 2011), elephants are allowed for greater flexibility in food choice as preferred and required. Therefore, a single type of food plant such as grass or a single species such as *Megathyrsus maximus* could not influence their body condition. The most preferred grasses exhibited lower nutritional quality than other preferred food plants, but the high water content in grass suggest that the moisture could influence the diet selection of the hindgut fermenting megaherbivore. While this preliminary study provides information on the diet composition of Sri Lankan elephants, further research should be conducted on the nutrition and food plants of the Sri Lankan elephant expanding across their large diet breadth, the varying seasons, and different localities of the elephant within the island. Additionally larger sample sizes and more in-depth analysis are needed to fully understand the nutritional contribution of different forage types and their implication for elephant health and well-being.

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Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23499–23506
ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)
https://doi.org/10.11609/jott.8438.15.7.23499-23506
#8438 | Received 16 March 2023 | Final received 10 May 2023 | Finally accepted 06 July 2023

COMMUNICATION

Does small mammal species richness have a bimodal elevation gradient in Sikkim Himalaya?

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Abstract: The most reported elevation gradients in species richness are a unimodal peak and linear decline. However, the overlap of different biogeographic realms in a region can influence such gradients. We used live-capture data on small mammals (voles, rats, mice, shrews, and pikas) to describe elevation gradients in species richness in Sikkim, where Afrotropical, Indo-Malayan, and Palearctic fauna occur in the lower, middle, and higher elevations, respectively. We sampled 38 trap lines in an elevation range of 300 m to 4,200 m, which we binned into nine elevation zones. Each trap line had 50 Sherman traps run for 3–5 nights during 2003–05 and 2012–13. We had a total of 9,069 trap nights with 430 captures, including 13 species of murid rodents, five ground shrews, two voles, and one each of pika and tree shrews. The capture rate in a trap line ranged from 0 to 19.7 per 100 trap night (mean = 5.30±0.767 SEM) with a peak at 2,501–3,001 m (3.29±0.644), coinciding with temperate broad leaf and conifer forests. Species richness seemed to have a minor peak at 501–1,000 m (2.50±0.645 species per trapline) and a clear peak at 3,001–3,500 m (3.29±0.644), coinciding with tropical forests and temperate mixed conifer forests, respectively. The apparent bimodal elevation gradient is due to the overlap of western Asian and Indo-Malayan fauna in the lower elevation and of the latter and Palearctic fauna in the higher elevation. More intensive sampling is needed to test this hypothesis that the overlap of biogeographic regions can influence elevation gradient in species richness.

Keywords: Altitude, capture rate, rodents, shrews, species composition.

Editor: Giovanni Amori, CNR-Research Institute on Terrestrial Ecosystems, Montelibretti, Italy. **Date of publication:** 26 July 2023 (online & print)

Citation: Khatiwara, S., J. Thapa & A. Kumar (2023). Does small mammal species richness have a bimodal elevation gradient in Sikkim Himalaya? *Journal of Threatened Taxa* 15(7): 23499–23506. https://doi.org/10.11609/jott.8438.15.7.23499-23506

Funding: Ministry of Environment, Forests and Climate Change, Government of India (no project fund number)Department of Biotechnology, Government of India (No.BT/01/NE/PS/NCBS/09)

Competing interests: The authors declare no competing interests.

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Author contributions: All authors participated in project design, JT and SK collected and analyzed data, AK administered the project, all authors participated in writing this manuscript.

Acknowledgments: We thank the Ministry of Environment, Forests and Climate Change, and Department of Biotechnology (both of Government of India) for funding this study; Department of Forests and Environment, Government of Sikkim, India, for providing research permits; Kalu Singh Rai, Sonam, Dawa Thendup, Andrew Chettri, and Jeewan Rai, for assistance in the field; Dr. Uma Ramakrishnan for the use of her laboratory; SK thanks Tanushree Srivastava for company and help on and off the field.

INTRODUCTION

The pattern of species richness along elevation gradient is among the most widely studied macroecology topics (Gaston 2000; McCain 2005; McCain & Grytnes 2010; Guo et al. 2013; Stevens et al. 2019). The most reported pattern is a unimodal mid-elevation peak followed by a monotonic decline in species richness with increasing elevation (Rahbek 1995; McCain & Grytnes 2010; Amori et al. 2019; Stevens et al. 2019). In Himalaya, mid-elevation peak has been reported in trees (Oommen & Shanker 2005; Acharya et al. 2011a), birds (Acharya et al. 2011b), amphibians (Chettri & Acharya 2020) and snakes (Chettri et al. 2010), although the elevation at which species richness peaks varies with the taxa. Species richness in lizards (Chettri et al. 2010) and butterflies decline linearly with elevation (Acharya & Vijayan 2015; Dewan et al. 2021). Nonvolant small mammals (primarily rodents and shrews) are perhaps the taxon in which elevation gradient in species richness has been most studied globally since this group is species-rich and locally abundant (Stevens et al. 2019). A mid-elevation peak is the most widely reported species richness pattern in non-volant small mammals (McCain 2005; McCain & Grytnes 2010; Stevens et al. 2019). However, the elevation gradient in species richness in small mammals has been little studied in the Himalayan region, in contrast with several studies in other parts of the world (see McCain 2005; Stevens et al. 2019 for reviews). Perhaps, the only study is Hu et al. (2017) who sampled small mammals in an elevation range of 1,800 m to 5,400 m on the southern slope of central Himalaya and reported a mid-elevation peak at 2,700–3,300 m, possibly a transition zone between Oriental and Palearctic regions.

The factors that influence elevation gradient patterns include climate (e.g., precipitation and temperature), space (e.g., species area richness and mid-domain effect), evolutionary history (e.g., speciation and extinction rates), and biological processes (e.g., competition, predation and habitat heterogeneity) (McCain & Grytnes 2010; Stevens et al. 2019). Although climatic factors have a major influence, climatic variables such as temperature and precipitation affect different taxa differently (Stevens et al. 2019). Most cold-blooded taxa show a decline in species richness with increasing elevation, since temperature declines with elevation. The factors that cause unimodal mid-elevation peak, widely reported in birds and mammals, are less known although water-energy balance (Hu et al. 2017) and productivity are possible factors (Stevens et al. 2019). Other factors such as species-area, evolutionary history and habitat heterogeneity have been studied even less (Stevens et al. 2019).

This paper examines elevation gradients in species richness in small mammals in Sikkim. Although the state of Sikkim in the eastern Himalaya is only 7,096 km^2 in area, it covers an elevation range of 200 m to >8,000 m. Sikkim also is uniquely located where the Indo-Malayan and Palearctic realms meet, and western Asian elements found in dry parts of India occur in the lower elevations. Among the small mammals reported from Sikkim (Naulak & Pradhan 2020), crocidurines (Dubey et al. 2008) and other Soricidae such as *Sorex* spp. and *Soriculus* spp. (Ohdachi et al. 2006), *Microtus* (Barbosa et al. 2018) are of Holarctic/Palearctic affiliation; *Rattus* (Robins et al. 2008) and the *Niviventer* (Ge et al. 2021) are of India-Malayan affiliation. Although taxa of Afrotropical affiliation are absent from those reported from Sikkim some are of West Asian origin, e.g., *Mus* (Suzuki et al. 2013) and *Tatera* (Khalid et al. 2022).

In this study, we examined the species richness patterns and composition of small mammal communities (murid rodents, pikas, ground, and tree shrews) along the elevation gradient from 230 m to 4,200 m. Our goal is to describe elevation gradients in species richness rather than to examine its relationship with several other factors reported in the literature (McCain 2005; Stevens et al. 2019).

Study area

Sikkim is a mountainous Indian state in the Himalayan biodiversity hotspot (Image 1), covering 7,096 km^2 and an elevation range from 200 m to ~8,000 m with an average slope of ~45˚ (Haribal 1992). Due to rugged terrain and rapid changes in elevation over short distances, temperature and precipitation vary considerably across the state. In southern Sikkim, the temperature varies from 6°C in winter to 35°C in summer, while winter temperature in the north falls much below freezing and the summer temperature is <20°C. Annual rainfall and precipitation days for 1995–96 was 1,310.44 mm and 91 at 300 m, 4,327 mm and 190 at 2,000 m, and 4,553.09 mm and 198 at 3,200 m (Krishna 2005). Almost the entire state of Sikkim comes in the catchment area of river Teesta.

The vegetation changes rapidly along the elevation gradient from the tropical semi-deciduous forest (<900 m) to tropical broadleaf (900–1,800 m), temperate broadleaf (1,800–2,800 m), temperate coniferous forest (2,800–3,800 m), sub-alpine (3,800–4,500 m), and alpine scrub to meadows (>4,500 m) (Haribal 1992).

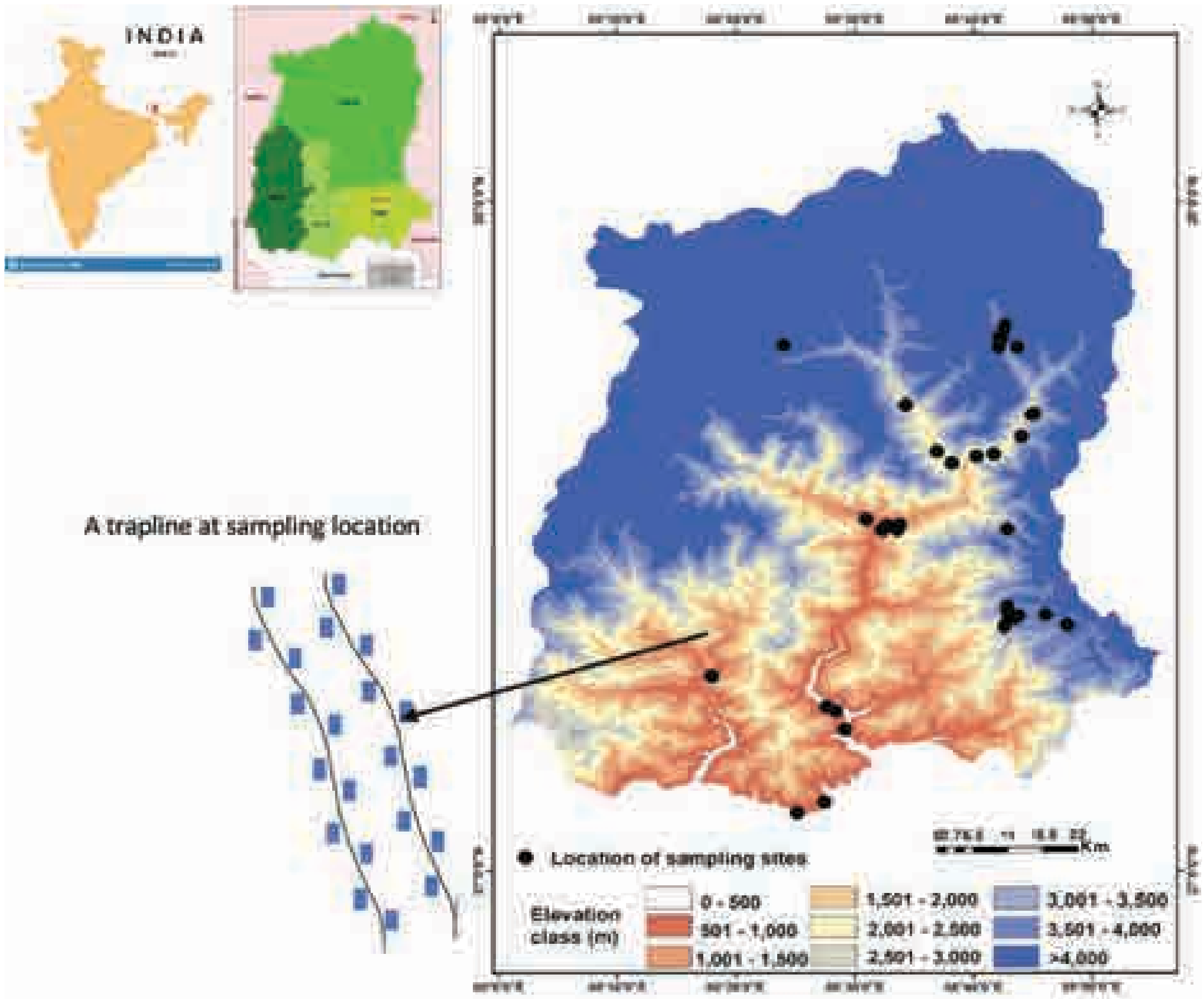

Image 1. State of Sikkim, showing administrative districts, 38 sampling locations, and a layout of traps along trail at each location.

The vegetation in the lower elevation mostly consists of *Shorea robusta, Terminalia myriocarpa, Pinus roxburgi*, and *Bombax ceiba* in the tropical semi-deciduous forest; *Engelhardtia spicata, Schima wallichii*, and *Castanopsis indica* in tropical broadleaf forest; *Quercus* sp., *Symplocos* sp., and *Rhododendron* sp. in the temperate broadleaf forest; *Abies densa*, *Juniperus recurva, Rhododendron* sp. in the coniferous forest; and dwarf *Juniperus* sp. and *Rhododendron* sp. mostly dominate the subalpine and alpine pastures of higher elevation areas in Sikkim. A more exhaustive vegetation classification identifies 12 forest types (Tambe et al. 2011). Some of the major forest types are the same as Haribal (1992) with similar elevation ranges, whiles others in Tambe et al. (2011) are subcategories within the major forest types in Haribal (1992).

METHODS

We sampled small mammals using Sherman live traps (7.5 × 9 × 23 cm) placed at 10 m intervals on alternate sides of existing natural trails in different elevation zones of the Sikkim Himalaya. We laid 38 traplines in an elevational range of 300 m to 4,200 m at an interval of ~500 m. We categorized this elevation range into nine elevation zones of 500 m, and sampled zones by laying three to seven traplines in each zone. Each such trap line had 50 traps which were run for three to five days, depending on the weather conditions. Since murid rodents, ground shrews and voles are mostly nocturnal, we kept the traps open only at night to prevent the capture of diurnal animals such as ground squirrels and birds. We checked and closed the traps every morning and baited them in the evening with a mixture of peanut butter, pulses, and crushed biscuits. The captured individuals were measured, weighed, photographed, ear punched (to detect recaptures) and released about 25 to 50 m away from the trap to minimize recaptures while also releasing the animals in the same vegetation type as they were captured. Species identification, in some cases up to the subspecies level, was done following on Agrawal (2000).

We located the sampling trails in forests that were least affected by human activities. Six trails (<800 m) in the south district, where agriculture (including fallow) covered about 30% of the land area, were in reserved forests as far away as possible from agricultural fields. Ten trails (>3,000 m) were in Kyongnosla Alpine Sanctuary, which had no human settlements and livestock grazing was prohibited. The remaining 22 trails (>1,000 m) were in protected areas and reserved forest in North Sikkim District where agriculture covered only 3% of the land area (http://slbcsikkim.co.in/General/Agriculture.aspx, accessed on 04 July 2023).

The uncertain and fluctuating temperature and precipitation profile of the study area allowed sampling only during certain months of the year. Thus, we did not sample the higher elevations (>2,000 m) in the winter months (November–April). The sampling in the north and south districts of Sikkim (Trapline No. 1–28) was from June 2003 to April 2004 and May 2005 to December 2005 (Thapa 2008) and that in East Sikkim District was done between May 2012 to June 2013.

Data Analysis

The capture rate for each trapline was calculated as (n/t_n) x100, where n is the animals trapped, and t_n is the number of trap nights. The number of species caught in each elevation zone was the observed species richness. Although this is always an underestimate of real species richness (Gwinn et al. 2015), we did not attempt to estimate the latter because both the number of trap lines and individuals caught were too few to meet the recommendations for the use of species richness estimators (Gotelli & Colwell 2010). Moreover, much of the underlying information needed for estimating species richness, such as species abundance distribution and detection probabilities (Gwinn et al. 2015) was unavailable. Therefore, we have used the number of species caught per trap line (of 50 traps) which can be considered the alpha diversity (McCain 2005) for examining the elevation gradient.

RESULTS

Elevational pattern of species

From over 9,069 trap nights of sampling effort, we live-trapped 430 individuals belonging to 22 taxa and 21 species (Table 1). The number of animals caught in a trapline varied from 0 (in four traplines in zone 1,001–1,500 m) to 46 (zone 2,501–3,000 m) with a mean of 11.32 (±1.742 SEM). We sampled only one elevation zone (3,501–4,000 m) in 2003–05 (n = 5 traplines) and 2012–13 (n = 2 traplines), which had similar capture rates per 100 trap nights (8.77 and 7.5, respectively). The capture rate in a trap line ranged from 0 to 19.7 (mean = 5.30±0.767). The capture rate was the highest at 2,501–3000 m, before declining, although still greater than at lower zones (Figure 1).

Muridae was the most species-rich family (13, including subspecies) in the region followed by Soricidae (ground shrews- including five species), Cricetidae (voles- including two species), Ochotonidae (pika), and Tupaiidae (tree shrew), the latter two families including one species each. The number of species captured in a zone was not significantly correlated either with the number of traplines (Spearman's rho = 0.527, p = .09), trap-nights (rho = 0.368, p = .330) or trapped animals (rho = 0.479, p = .192). However, zone 3,001–3,500 m accounted for the highest number of trapped animals (114) and species richness corresponding to the maximum effort in the zone with 1,661 trap-nights in seven traplines (Table 1).

Species richness per trapline had a minor peak at 500–1,000 m and a major peak at 3,000-3,500 m (Figure 1). The differences in capture rate and species richness among the five vegetation types was similar to the elevation gradient (Figure 2). The capture rates were highest in the subalpine and conifer forests and lowest in the tropical forests at the lower elevations. Species richness per trapline appeared to show two peaks: a small peak in the tropical deciduous forest and a larger peak in the subalpine forest.

Species composition

The species richness (including subspecies) in an elevation zone ranged from three to eight, the composition of which changed from lower to higher elevation (Figure 3). Three species of *Mus* occurred primarily in the lower elevations (<2,000 m), while five species (*Microtus sikimensis, Ochotona* sp., *Pitymys* sp., and *Sorex* sp.) occurred primarily at >3,000 m, while *Soriculus nigrescens* occurred >1,000 m. The remaining 12 species had narrow elevation ranges (e.g.,

Table 1. Details of trapping effort and captures of small mammals in nine elevation zones in Sikkim.

Elevation Zone (in m)	N of trap-lines	N trap nights	N of animals	N of taxa in zone	Taxa trapped (see below for taxa identities)
<500	3	794	9	3	4, 14, 17
501–1000	4	1171	36	6	1, 4, 5, 8, 17, 22
1001–1500	7	1449	44	4	2, 4, 17, 20
1501–2000	3	568	13	5	6, 8, 15, 16, 18, 20
2001–2500	3	741	29	4	8, 10, 15, 20
2501–3000	3	460	57	4	2, 7, 8, 20
3001–3500	7	1661	114	8	2, 3, 7, 11, 12, 13, 19, 20
3501–4000	5	1475	93	5	1, 3, 9, 11, 19
4001–4500	3	750	35	3	1, 3, 11

1. *Crocidura* sp.
2. *Episoriculus caudatus*
3. *Microtus sikimensis*
4. *Mus mus castaneus*
5. *Mus mus homurus*
6. *Mus pahari*
7. *Niviventer eha*
8. *N. fulvescens*
9. *Niviventer* sp.
10. *N. niviventer*
11. *Ochotona* sp.
12. *Pitymys* sp.
13. *Rattus blandfordi*
14. *R. nitidus*
15. *R. r. brunne*
16. *R. r. tistae*
17. *R. sikkimnesis*
18. *R. turkestanicus*
19. *Sorex* sp.
20. *Soriculus nigrescense*
21. *Suncus murinus*
22. *Tupia* sp.

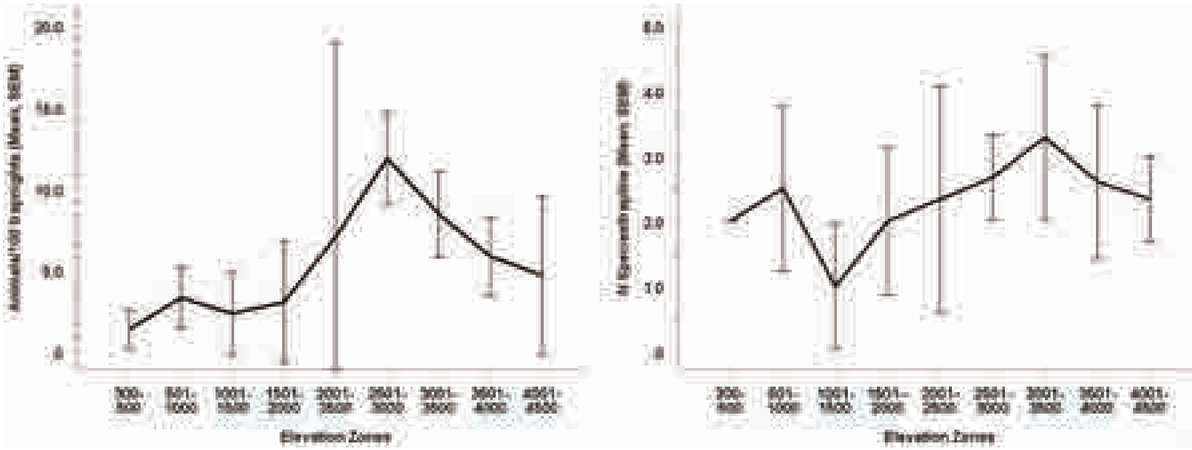

Figure 1. Changes in total capture rates (left) and number of species of small mammals caught per trap line in nine elevation zones (right).

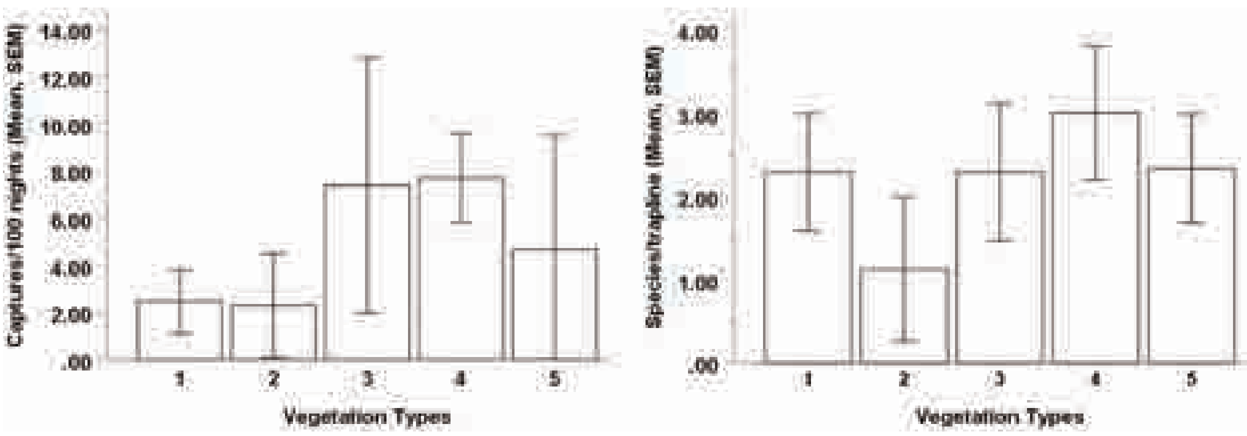

Figure 2. Capture rates (left) and number of species of small mammals caught per trapline (right) in five vegetation types in Sikkim: 1—Tropical dry deciduous | 2—Tropical broadleaf | 3—Temperate broadleaf | 4—Temperate mixed coniferous | 5—Subalpine.

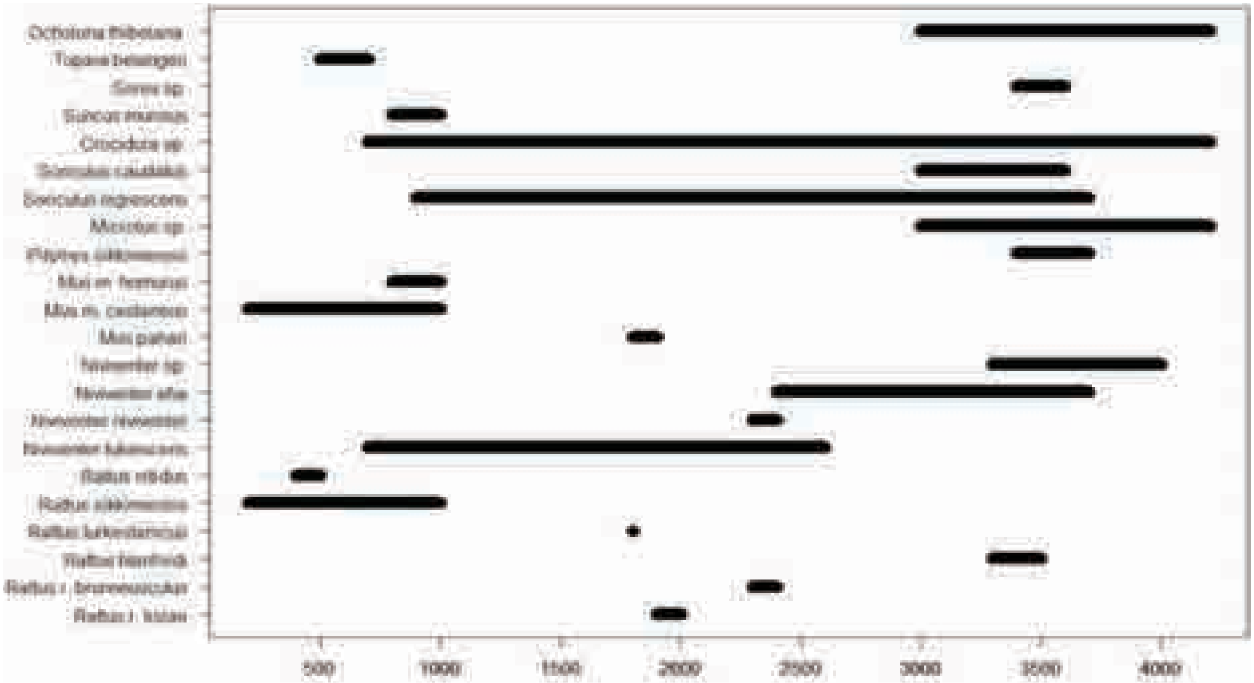

Figure 3. Elevation ranges of 22 species of small mammals caught during 2003–05 and 2012–13.

Tupaia sp.). Only *Crocidura* sp. and *Soriculus caudatus* had wide elevational ranges (Figure 3). This pattern indicates a type of substitution of rats and mice in the low and mid elevations by voles and ground shrews at the higher elevations. At the family level, Muridae and Soricidae were captured from all elevation zones except >4,000 m for the former and <500 m for the latter. Ochotonidae and Cricetidae were captured only above 3,000 m.

DISCUSSION

We used data from live trapping of small mammals in an elevation range from 230 m to 4,200 m in Sikkim to describe the elevation gradient in species richness and compositional changes. The capture rates were greater in the higher elevations, although there was considerable variation within each elevation zone. While some studies have reported higher capture rates in higher elevations (e.g., Rickart et al. 1991; Heaney 2001), others have reported lower capture rates (e.g., Li et al. 2003). An important factor influencing capture rates is the sampling season since small mammals show drastic seasonal fluctuations in abundance, especially in higher elevations. However, we sampled lower elevations (<2,000 m) after summer showers in March up to September, and the higher elevations during June to early November, when small mammal abundances were expected to peak. Therefore, at their respective peaks, abundances are greater in the higher elevations. Similarly, abundances in subalpine forests are far greater than in the tropical forests in the lower elevation. The capture rates of <4% in the tropical forests in this study is comparable to that reported from undisturbed rainforests in the Western Ghats – 2.12% (Kumar et al. 2002) and 4.38% (Kumar et al. 1997) although less than reported from sites in tropical Africa (6.88%, Hounmavo et al. 2023). Capture rates in temperate forests are often much greater and sometimes very high depending on fruit masts (Grendelmeier et al. 2018).

Species richness showed a clear peak at 3,001–3,500 m, coinciding with mixed conifer forest, and a smaller peak at 501–1,000 m, coinciding with tropical forests (deciduous and broadleaf). In unimodal richness gradients, the peak occurs at higher elevations in taller mountains (McCain 2005) like the larger peak in this case. However, we believe that on biogeographical considerations, two peaks are likely. In the lower elevations, taxa of West Asian and Indo-Malayan affiliations overlap at the edge of their respective elevation ranges and at the sub-alpine forests where taxa of Indo-Malayan and Palearctic affiliations overlap. Out of the 56 studies that McCain (2005) reviewed, only two had peaks in alpha diversity at lower and higher elevations, perhaps due to a lack of sampling of the entire gradient or in mid-elevations. This was probably not the case in our study since we had among the highest number of traplines (seven) in 1,001–1,500 m, which had the lowest species richness. Human alteration of habitat was not a factor since these seven trails were in protected forests in North Sikkim District, where agricultural land is only 3%, and human population density was 10 per km^2 (www.indiacensus.net/states/sikkim accessed on 04 July 2023).

In the same landscape, trees show a unimodal peak at ~1500 m, coinciding with tropical broadleaf forests (Acharya et al. 2011a). Total species richness in amphibians also peak at the same elevation (Chettri & Acharya 2020), whereas reptile species richness peak at 500–1,000 m, coinciding with tropical deciduous forests, although lizards decline linearly with elevation and snakes show a unimodal peak (Chettri et al. 2010). Peak in the bird species richness at 1,800–2,000 m (Acharya et al. 2011b), overlapped with temperate broad leaf forests. Overall species richness in butterflies declines linearly with elevation (Dewan et al. 2021). Our data show a clear peak in small mammal species richness at a higher elevation (3,001–3,500 m) compared to the above taxa in Sikkim. This is due to the presence of species of Palearctic/Holarctic affiliation in Cricetidae (Dubey et al. 2008; Barbosa et al. 2018), Soricidae (Ohdachi et al. 2006) and Ochotonidae (Melo-Ferreira et al. 2015), along with species of Indo-Malayan affinity, e.g., *Niviventer* spp. (Ge et al. 2021). In Gyirong Valley in Central Himalaya, Hu et al. (2017) reported 22 species (from 21,600 trap nights) with similar species composition (13 Muridae, 3 Cricetidae, 3 Soricidae, and 3 Ochotonidae). The species richness peaked at 2,700–3,300 m, covered by mixed conifer and subalpine forests (Liang et al. 2020). In our study, the species richness peaked at 3,001–3500 m, where the same forest types occur. Hu et al. (2017) suggested that the peak species richness was probably due to the overlap of Indo-Malayan and Palaearctic regions, although they did not examine species composition in this context. Our data also suggests a smaller peak at 501–1,000 m, due to the presence of species rich Indo-Malayan taxa such as *Rattus* (Robins et al. 2008) and *Niviventer* (Ge et al. 2021), along with species of West Asian affinity such as *Mus* (Suzuki et al. 2013). Hu et al. (2017) did not include forests at <1,800 m with tropical deciduous and broadleaf forests, where western Asian and Indo-Malayan fauna overlap. This overlap can result in another peak in species richness, as our study shows. Thus, Himalaya in Sikkim probably has a bimodal peak in alpha species richness of small mammals. Only a study with more intensive trapping effort can test this hypothesis.

CONCLUSIONS

We examined the elevation gradient in species richness of small mammals using data from live traps covering an elevation range of 230–4,200 m. There is a clear peak in species richness at 3,001–3,500 m and probably another minor peak in the lower elevation (501–1,000 m). These peaks are likely because of the overlap of West Asian and Indo-Malayan fauna in the lower elevation and of the latter and Palaearctic fauna in the higher elevation. This bimodal peak contrasts with unimodal peaks reported from the area in plants, amphibians, snakes, and birds and linear decline reported in lizards and butterflies. Most of the reports of unimodal peaks in small mammals come from areas where biogeographic realms do not overlap, or this issue has not been addressed. The Himalaya in Sikkim is an ideal site to examine the influence of overlaps of biogeographic realms on elevation gradients.

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Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23507–23513
ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)
https://doi.org/10.11609/jott.8411.15.7.23507-23513
#8411 | Received 17 February 2023 | Final received 30 May 2023 | Finally accepted 01 July 2023

OPEN ACCESS

COMMUNICATION

Re-sighting record of Kelaart's Pipistrelle *Pipistrellus ceylonicus* (Kelaart, 1852) (Mammalia: Chiroptera: Vespertilionidae) from Rajasthan, India

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Abstract: Despite numerous studies and surveys targeting Chiroptera in Rajasthan since 1955, *Pipistrellus ceylonicus* has not been observed in the state for more than a century since 1913. Based on an adult male specimen recovered from Kusthala village in Sawai Madhopur district of Rajasthan, we report the occurrence of this species from the state after more than a century.

Keywords: Bat, occurrence, population, rediscovery, Sawai Madhopur,

Hindi: वर्ष 1955 से राजस्थान में काईरोप्टेरा से जुड़े कई अध्ययनों और सर्वेक्षणों के बावजूद, 1913 के बाद से, यानी लगभग एक सदी से भी अधिक समय से राज्य में पिपिस्ट्रेलस सीलोनिकस नहीं देखी गयी है। राजस्थान के सवाई माधोपुर जिले के कुस्तला गांव से प्राप्त एक वयस्क नर चमगादड़ नमूने के आधार पर, हम इसे रिपोर्ट करते हैं, जो एक सदी से भी अधिक समय के बाद राज्य में इस प्रजाति को देखा गया है।

Editor: P.O. Nameer, Kerala Agricultural University, Thrissur, India. **Date of publication:** 26 July 2023 (online & print)

Citation: Khandal, D., D.L. Bohra & S.S. Talmale (2023). Re-sighting record of Kelaart's Pipistrelle *Pipistrellus ceylonicus* (Kelaart, 1852) (Mammalia: Chiroptera: Vespertilionidae) from Rajasthan, India. *Journal of Threatened Taxa* 15(7): 23507–23513. https://doi.org/10.11609/jott.8411.15.7.23507-23513

Funding: Tiger Watch.

Competing interests: The authors declare no competing interests.

Author details: Dr. Dharmendra Khandal has served as conservation biologist with Tiger Watch since 2003. His work with Tiger Watch has involved pioneering ground breaking initiatives in proactive anti-poaching, the monitoring of wildlife & scientific research. He has also forged new frontiers in the world of community based conservation through the Village Wildlife Volunteer program in the Ranthambhore Tiger Reserve. He is also the co-author of Unexplored Ranthambhore, a first of its kind book on the canids and Striped Hyena in Ranthambhore. Dr. Dau Lal Bohra, is currently head of the Department of Zoology at the Seth Gyaniram Bansidhar Podar College in Jhunjhunu, Rajasthan. He has numerous research papers to his credit and is recognised for his significant contributions to vulture conservation in Rajasthan. Dr. S. Talmale, is a taxonomist working on Indian small mammals and (Insecta) Odonata with several research papers and books to his credit. He is currently affiliated with the Zoological Survey of India.

Author contributions: This bat was spotted and captured in an injured state by DK. Unable to rehabilitate it, after its natural death, its skull was removed by DLB for the identification of the specimen. The morphometric measurements were also carried out by him. SST identified this specimen and prepared the draft of this paper.

Acknowledgements: The authors are grateful to Mr. Dieter Gutmann & Mrs. Liz Gutmann, and Tiger Watch for their unstinting support.

INTRODUCTION

A small-sized bat *Pipistrellus ceylonicus* was initially recorded from Rajasthan at Mt. Abu during the mammal surveys of India, Burma, and Ceylon in 1911–1923 conducted by the Bombay Natural History Society (BNHS). In this survey two males and one female of this species were collected in March–July 1913 (Ryley 1914). Since that time, a large number of surveys targeting the chiropteran fauna of the state were undertaken e.g., Garg (1955); Prakash (1961, 1963, 1973); Agrawal (1967); Biswas & Ghosh (1968); Sinha (1975, 1976a,b, 1977, 1978, 1979, 1980a,b, 1981, 1983, 1996); Gaur (1981); Advani (1982); Ramaswami & Kumar (1963); Kumar (1965); Wason (1978); Agarwal & Gupta (1982); Lall (1985); Bhupathy (1987); Gupta & Trivedi (1989); Trivedi & Lall (1989); Sharma (1986); Agarwal et al. (1981); Trivedi (1991); Purohit & Senacha (2002, 2004a,b); Senacha (2003, 2006); Trivedi et al. (2003); Dookia (2004); Dookia & Tak (2004); Senacha & Purohit (2004); Trivedi & Lall (2004, 2006); Senacha et al. (2006); Srinivasulu & Srinivasulu (2006); Purohit et al. (2006); and Khandal et al. (2022). However, *Pipistrellus ceylonicus* was not recorded in any of these surveys (Figure 1).

MATERIAL AND METHODS

In November 2021, an injured adult male *Pipistrellus* was rescued from Kusthala village (25.9694^0N, 76.2929^0E) in Sawai Madhopur, Rajasthan (Image 1 & 2). The bat was treated at home and kept in a box but did not survive. The specimen collection site is near the state highway close to the village of Kusthala, in the district of Sawai Madhopur. The landscape is dominated by agricultural fields close to a small human settlement. The area lies near a very significant ecosystem, i.e., the forests of Ranthambhore Tiger Reserve which is barely 4.5 km away. Specimen and habitat photographs were taken with a Nikon D850 DSLR equipped with a 17–35 mm lens. Morphological data was taken by manual examination in which measurements were taken with a digital caliper.

Figure 1. Map showing the new and old distribution localities for Kelaart's Pipistrelle in Rajasthan state.

Image 1. Portrait of Kelaart's Pipistrelle *Pipistrellus ceylonicus* (Kelaart, 1852) (present study)

Image 2. Close up of Kelaart's Pipistrelle *Pipistrellus ceylonicus* (Kelaart, 1852) (present study)

Table 1. Morphological, cranial and dental measurements of *Pipistrellus ceylonicus* (Kelaart, 1852) (all measurements are in millimeters)

	Measurement (mm)	Bates & Harrison (1997)	Korad & Yardi (2004) (n=7)	Present study (n=1)
1	Head and body length (HB)	45.5 - 64.0	46-51.4	41.2
2	Tail length (T)	30.0 - 45.0	29-38.5	31
3	Hind foot length, including claw (HF)	6.0 - 11.0	6-8.5	8.4
4	Forearm length (FA)	33.0 - 42.0	35-38.2	39
5	Wingspan (WSP)	227-262	227-252	243
6	5th Metacarpal length (5MT)	30.7 - 36.7	33.0-34.5	33.7
7	4th Metacarpal length (4MT)	32.6 - 38.5	34.4-35.8	34.8
8	3rd Metacarpal length (3MT)	33.0 - 39.0	34.5-36.4	33.1
9	Ear length (E)	9.5 - 14.0	9.5-14	11.2
10	Tibia length (Tb)	NA	13.5-15.0	14.1
11	Greatest length of skull (GTL)	14.4 - 15.8	13.5-15.5	14.9
12	Condylocanine length (CCL)	13.1 - 14.3	13.0-14.0	13.6
13	Zygomatic breadth (ZB)	9.2 - 11.0	9.0-10.0	9.2
14	Breadth of braincase (BB)	6.8 - 7.8	7.7-8.0	7.1
15	Postorbital constriction (PC)	3.7-4.3	3.8-4.5	3.9
16	Maxillary toothrow length (CM^3)	5.2 - 5.9	5.4-6	5.8
17	Mandibular toothrow length (CM_3)	5.7 - 6.5	5.6-6.6	6.2
18	Width across third molars (M^3-M^3)	6.2 - 7.2	6.6-7.8	6.8
19	Mandible length (M)	10.6 - 12.0	10.6-11.6	10.9
20	Width of rostrum (RW)	5.7-7.1	5.5-7.0	5.9

The specimen was preserved in 70% ethanol. Standard morphological measurements of the specimen and cranio-dental measurements of the extracted skull were taken using a digital calliper accurate to the nearest 0.1 mm and 0.01 mm, respectively. The morphological and craniodental description (Table 1) of the bat matched with descriptions provided by Bates & Harrison (1997) and Korad & Yardi (2004) confirming the specimen as *Pipistrellus ceylonicus* (Kelaart, 1852).

RESULTS AND DISCUSSION

Kelaart's Pipistrelle, *Pipistrellus ceylonicus* is a large sized *Pipistrellus* with a forearm length of 33–42 mm (Bates & Harrison 1997). They have variable dorsal pelage coloration ranging from grey-brown to chestnut, reddish or golden-brown colour. The ears, naked areas of the face, wings and interfemoral membrane are a uniform dark brown. The present specimen was grayish-brown dorsally and had dark hairs with pale grey tips on the ventrum (Image 1 & 2). The skull is robust with condylo-canine length of 13.6 mm and the upper toothrow length (cm^3) is 5.8 mm (Image 3 A & B). The first upper incisor (i^2) is bicuspidate; the second incisor (i^3) is larger in size and two-thirds the height of i^2. The first small premolar (pm^2) intruded into the toothrow, and was not visible on the outside (Image 4 A & B). The upper canine and posterior premolar (pm^4) are almost in contact. The lower incisors are trifid and overlap slightly (Image 5).

Three subspecies under *P. ceylonicus* recognized from India by Ellerman & Morrison-Scott (1951), viz., *Vesperugo indicus* Dobson, 1878 (type from Mangalore, Malabar Coast, Karnataka), *Pipistrellus chrysothrix* Wroughton, 1899 (type from Mheskatri, Surat Dangs, Gujarat) and *P.c. subcanus* Thomas, 1915 (type from Yalala, Junagarh, Kathiawar, Gujarat). Individual body color variation was observed in individuals of the same colony of *P. ceylonicus* by Brosset (1962). Based on variation in colour, Khajuria (1978, 1980) has synonymised *crysothrix* with *indicus.* Lal (1984) has considered both *crysothrix* and *subcanus* as synonyms of *Pipistrellus cylonicus indicus.* Moratelli & Burgain (2019) considered all populations of *P. ceylonicus* from the mainland Indian subcontinent with distribution in eastern and southeastern Pakistan, India and Bangladesh are to represent a single subspecies,

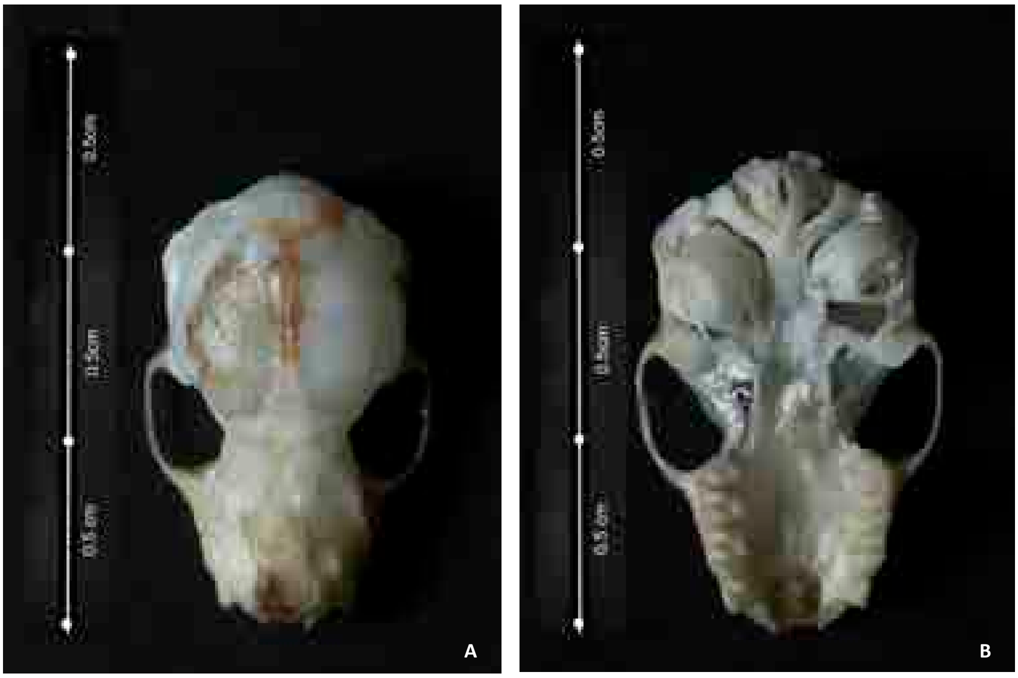

Image 3. *Pipistrellus ceylonicus* skull: A—Dorsal view| B—Ventral view. © Dharmendra Khandal.

Image 4. *Pipistrellus ceylonicus*, skull: A—Lateral view | B—Front view. © Dharmendra Khandal.

Image 5. *Pipistrellus ceylonicus,* lower jaw with dental arrangement. © Dharmendra Khandal.

Pipistrellus ceylonicus indicus Dobson, 1878.

Some of the earlier works on taxonomy, biology and ecology of bats of Rajasthan (Prakash 1961; Agrawal 1967; Biswas & Ghosh 1968; Sinha 1976a,b, 1978, 1980a,b) did not report any new occurrence data of *P. ceylonicus* from the state. Ghosh (2008), while preparing a catalogue of bats specimens available in the National Zoological Collection at Zoological Survey of India, Kolkata, mentioned the distribution of the species in Rajasthan based only on the past record by Ryley (1914) and without any new collection data.

In view of its widespread distribution and adaptable nature, IUCN Red List categorized the species as 'Least Concern' (LC) (Srinivasulu & Srinivasulu 2019). It is apparently of rare occurrence and extensive surveys are needed to determine the status of the species in the state.

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Threatened Taxa

Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23514–23520

OPEN ACCESS

ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)

https://doi.org/10.11609/jott.8415.15.7.23514-23520

#8415 | Received 20 February 2023 | Final received 25 April 2023 | Finally accepted 16 July 2023

COMMUNICATION

An assessment of the diet of Brown Fish-Owl *Ketupa zeylonensis* (J.F. Gmelin, 1788) (Aves: Strigiformes: Strigidae) from two localities in the foothills of the Western Ghats of Goa, India

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Abstract: The Brown Fish-Owl *Ketupa zeylonensis* is a large nocturnal bird of prey that has a vast distribution range. However, there is a significant literature gap on the ecology of this species in the Western Ghats ecoregion, particularly in regard to its food spectrum. In the present study, we assessed the diet composition of this species in the foothills of the Western Ghats of Goa, India. The diet was evaluated by analysing the undigested prey remains in regurgitated pellets obtained from the banks of forest streams and roosting sites. A total of 104 pellets were collected from two localities that exhibited similar landscape characteristics. Our analysis indicated that crabs contributed to a significant proportion of the diet of the species (75.47%), followed by amphibians (frogs, 8.02%), fishes (7.08%), reptiles (snakes, 2.83%), birds (2.36%), scorpions (1.89%), and insects (Odonata, 0.47%). Additionally, 1.89% (n = 4) of the prey items could not be identified due to their disintegrated nature. Furthermore, an assessment of Food Niche Breadth (FNB) indicated that *K. zeylonensis* exhibited a high degree of specialization in terms of its diet in the study areas.

Keywords: Diet analysis, feeding ecology, food niche breadth, food spectrum, forest streams, owl pellets, prey composition, relative frequency of occurrence.

Abbreviations: CITES—Convention on International Trade in Endangered Species of Wild Fauna and Flora | FNB—Food Niche Breadth | IUCN—International Union for Conservation of Nature | NP—National Park | RFO%—Relative Frequency of Occurrence | UNESCO—United Nations Educational, Scientific and Cultural Organization | WS—Wildlife Sanctuary.

Editor: Prachi Mehta, Wildlife Research and Conservation Society (WRCS), Pune, India. **Date of publication:** 26 July 2023 (online & print)

Citation: Dias, S.J. & A.S. Borker (2023). An assessment of the diet of Brown Fish-Owl *Ketupa zeylonensis* (J.F. Gmelin, 1788) (Aves: Strigiformes: Strigidae) from two localities in the foothills of the Western Ghats of Goa, India. *Journal of Threatened Taxa* 15(7): 23514–23520. https://doi.org/10.11609/jott.8415.15.7.23514-23520

Funding: No funding was acquired to carry out the study.

Competing interests: The authors declare no competing interests.

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Author contributions: SJD & ASB: Conceptualized the study and carried out the field work; SJD: Performed the data analysis; SJD & ASB: Prepared the draft of the manuscript.

Acknowledgements: We are grateful to the trustees of Planet Life Foundation and the staff of Nature's Nest Nature Resort for their support during the project. Our gratitude to the Goa State Biodiversity Board for granting the necessary approvals for the project. Special thanks to Swanand Patil of Arcane Conservancy for his assistance during the initial stages of the project and for providing us with the photograph of Brown Fish-Owl from his personal collection. We would also like to thank Rohan Gaonkar and Gautam Gad for extending their support. We are thankful to Sophia Dias, Anselia Da Costa, and Dr. Claret Shastry for proofreading the manuscript. Lastly, we would like to appreciate the efforts of the reviewers for their constructive comments and suggestions on the manuscript.

INTRODUCTION

Birds of prey occupy the apex position in food web assemblages. Therefore, they are considered to be important bioindicators of the environments in which they persist (González-Rubio et al. 2021). The taxonomic order Strigiformes is represented by 250 extant species of owls distributed across the world (Gill et al. 2023). This order is divided into two families: (i) Tytonidae, which includes barn owls, bay owls, and grass owls, and (ii) Strigidae, which includes true (or typical) owls (Sieradzki 2023). India is home to 32 species of owls, 13 of which are found in the state of Goa (Baidya & Bhagat 2018; BirdLife International 2020). The Brown Fish-Owl *Ketupa zeylonensis* is a nocturnal bird of prey that is distributed across southern and southeastern Asia with isolated populations occurring in Turkey and Iran, and vagrant populations occurring in Seychelles (Birdlife International 2016). It is a large bird (approx. 56 cm) having bright yellow eyes and outward-facing ear tufts. It exhibits rufous-brown upper parts with heavy streaking, and pale underparts with dark streaks (Ali 2002; Kazmierczak & Perlo 2012; Grewal et al. 2016). The species is classified as 'Least Concern' in the IUCN Red List of Threatened Species. Although global populations of this species have not been evaluated, it is suspected to be in decline due to habitat destruction (Birdlife International 2016). In addition, the species is listed under 'Schedule I' of the Indian Wild Life (Protection) Amendment Act, 2022 and under Appendix II of CITES (Ministry of Law and Justice 2022; CITES 2023). In India, this species faces threats from the illegal wildlife trade, persecution by fishermen, and its use in witchcraft (Ahmed 2010).

The Brown Fish-Owl inhabits deciduous, semi-deciduous and evergreen woodland ecosystems and is found in close proximity to water bodies. Its diet is reported to constitute crabs, fish, frogs, reptiles, birds, mammals, and carrion (Ali 2002; Bindu & Balakrishnan 2015; Grewal et al. 2016).

Owls are highly specialized hunters that regurgitate undigested prey remains such as bones, feathers, hair, scales, and other exoskeletal structures of their prey in the form of compact pellets. The analysis of regurgitated pellets has proven to be a robust technique to assess the food spectrum of owls and understand the diversity and population structure of prey species (Meek et al. 2012; Andrade et al. 2016). In an Indian context, published literature on the diet composition of the Brown Fish-Owl is sparse. Vyas et al. (2013) reported the food spectrum of *K. zeylonensis* from Jambughoda WS in Gujarat. However, there is a significant literature gap in the diet composition of the species from the Western Ghats ecoregion, particularly in the context to the Indian state of Goa. This study was carried out to understand the diet composition of the species in two sites located in the foothills of the Western Ghats of Goa.

MATERIALS AND METHODS

Study Area

Goa is located on the western coast of India (15.492°N, 73.826°E) (Figure 1). The Western Ghats is a 1,600 km long mountain range that runs parallel to the western coast of the Indian peninsula and extends through the Indian states of Gujarat, Maharashtra, Goa, Karnataka, Tamil Nadu, and Kerala. These mountains are recognized as one of the world's eight 'hottest hotspots' for biological diversity and endemism (Molur et al. 2011; UNESCO 2023). In Goa, these mountains pass through the eastern regions of the state where a significant section of the range is protected through four protected areas: Mhadei WS, Bhagwan Mahavir WS & NP, Netravali WS, and Cotigao WS. The vegetation type of the Western Ghats of Goa is varied and includes tropical evergreen, semi-evergreen, and moist mixed deciduous forests (Goa Forest Department 2023). This study was conducted along forest streams that originate from the Western Ghats. The sections of the streams surveyed for this study were located outside the boundaries of protected areas. Study Area 1 was located near Mhadei WS and Study Area 2 was located near Bhagwan Mahavir WS & NP. The streams that were considered for the study were of the perennial and intermittent type. The general vegetation type of the study areas is dominated by tropical evergreen, semi-evergreen, and moist mixed deciduous forests. In addition, both study areas were located in close proximity to plantations and human settlements. The streams considered for this study are part of a larger catchment system that empties into the Mahadayi River of Goa (see Figure 1). The aerial distance between the two study areas was approximately 16.7 km.

Data Collection

This study was conducted from 20 October 2022 to 5 February 2023. Prior to this study, Brown Fish-Owl activity in Study Area 1 was established by conducting field surveys. In addition, the feeding and breeding activity of this species in Study Area 2 was recorded for over two years with the help of camera traps and

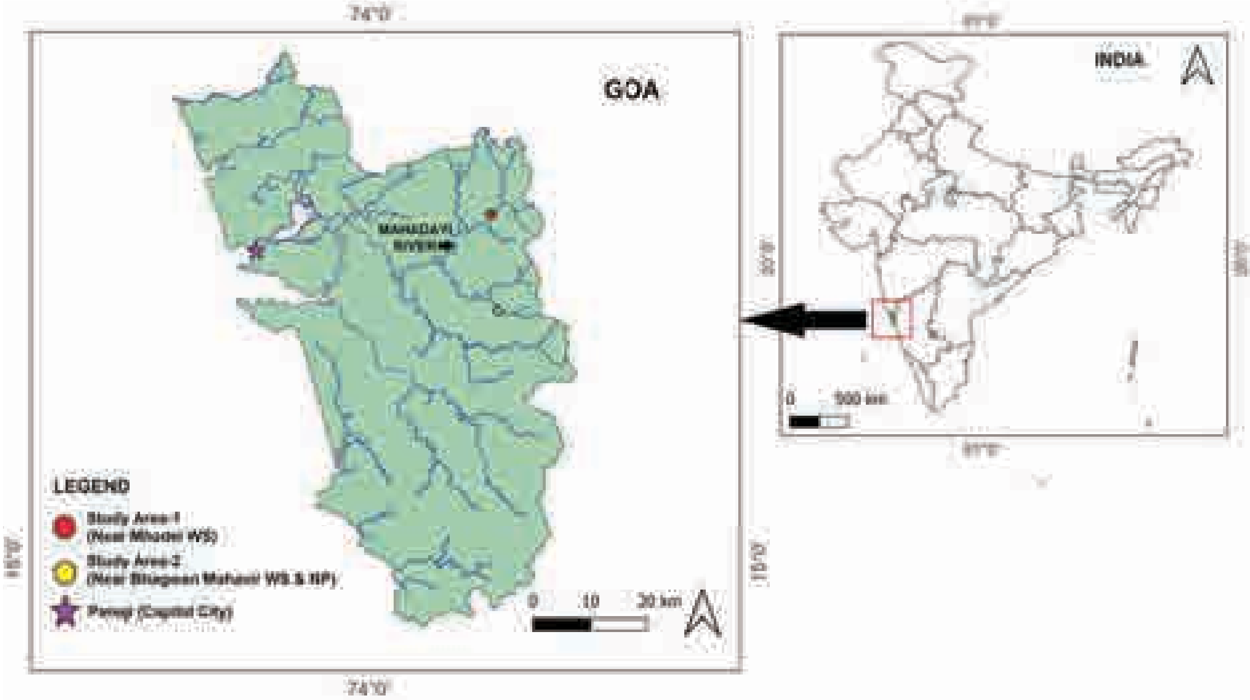

Figure 1. Map depicting the study areas. WS—Wildlife Sanctuary | NP—National Park.

direct observations respectively. This was part of a larger nocturnal wildlife monitoring effort by Planet Life Foundation, Belloy, Nuvem, Goa and Nature's Nest Nature Resort, Surla, Sancordem, Goa. Brown Fish-Owl pellets were collected from the study areas once a week. The pellets were usually found deposited along stream banks and in close proximity to roosting sites (Image 1). A total of four roosting sites were identified across our study areas based on repetitive pellet deposition observed during our surveys. The entire pellet was collected and temporarily stored in plastic zip-lock bags. Prior to analysis, we manually removed all conspicuous debris from the pellet by hand. Following this, the pellets were soaked in 70% ethyl alcohol for 24 h to kill all microorganisms. The pellets were then air-dried for 24 h to remove moisture. During analysis, the dry weight of each pellet was recorded using a weighing balance with 0.001 g accuracy. The prey items in the pellets were then sorted into eight categories: crabs, insects, scorpions, fishes, amphibians, reptiles, birds, and unidentified prey. These prey categories were established based on literature review and field observations. As we did not have access to reference specimens, the items in the pellets could not be identified at the species level. Identification of the prey items was carried out using reference books and taxonomic keys (Verma 2014; Ganguly et al. 2015; Saxena & Saxena 2019; Mehta et al. 2020; Mishra et al. 2021). As most of the items in the pellets were conspicuous, identification and sorting were possible by the naked eye. However, we used a compound microscope (ESAW SM-02, ESAW India, Ambala Cantt, Haryana, India) set at 10x magnification to identify the inconspicuous prey remains. Arthropods were identified primarily from structures such as mouthparts, chelipeds, pereiopods, abdomen, and carapace (in the case of crabs); wings (in the case of insects); pedipalps, cephalothorax shield, mesosoma, metasoma, walking legs, and telson (in the case of scorpions). Scorpion identification was also aided by shining an ultraviolet light at 395 nm and observing fluorescence (Gaffin et al. 2012) (Image 1). Chordates were identified from endoskeletal and exoskeletal structures such as bones and scales (in the case of fishes), bones and mouthparts (in the case of amphibians i.e., frogs), vertebrae, ribs, and skin (in the case of reptiles i.e., snakes), and bones and feathers (in the case of birds). The prey items that could not be identified were sorted into the 'unidentified' category. For each pellet, we estimated the number of individuals for each prey category (Table 1). The data for both study sites was pooled and subsequently analyzed.

Data Analysis

We estimated the Relative Frequency of Occurrence (RFO%) for each prey group by dividing the number of occurrences of each prey category by the total number of occurrences of all prey categories multiplied by 100 (Mehta et al. 2020). To assess the diversity of prey in the owl diet, we estimated the Food Niche Breadth (FNB) by employing the standardized Levin's Index (B_A) formula (Levins 1968; Colwell & Futuyma 1971; Mehta et al. 2018) as follows:

$$B_A = \left(\frac{1}{\sum P_i^2} - 1\right) \times \frac{1}{n-1}$$

Where P_i is the proportion of i^{th} prey category and n is the number of prey categories recorded in the diet of the Brown Fish-Owl. This standardized index computes a value that can range from 0–1. Values closer to 0 indicate a specialist diet whereas values closer to 1 indicate a generalist diet (Mehta et al. 2018).

RESULTS

A total of 104 Brown Fish-Owl pellets were collected during the present study (50 pellets from Study Area 1 and 54 pellets from Study Area 2). The average dry weight of the pellets was estimated to be 4.053 g (SD = ± 2.627; Range = 0.590–12.953). The total number of prey individuals recorded was 212. The average number of prey individuals per pellet was estimated to be 2.029 (SD ± 1.074; Range = 1–5). The diet of the Brown Fish-Owl was dominated by crabs followed by amphibians (frogs), fishes, reptiles (snakes), birds, scorpions, and insects (Odonata). The unidentified prey individuals constituted a minor portion of the diet (n = 4, Table 2). Although we were unable to positively identify the type of prey items in the 'unidentified' category due to their disintegrated nature, we were able to identify the remnants as vertebrates. In such cases, all the unidentified remains having similar characteristics were assumed to originate from a single individual. The number of occurrences of prey categories was largely comparable across the two study areas. However, insects were only present in the pellets collected from Study Area 1 (Odonata, n = 1) and scorpions were only present in pellets collected from Study Area 2 (n = 4) (Figure 2). Lastly, the Food Niche Breadth (FNB) value was estimated to be 0.1, indicating that the Brown Fish-Owl exhibits a high degree of specialization in terms of its diet in the study areas. The diet composition of the species in the present study has been detailed in Table 2.

DISCUSSION

The Brown Fish-Owl is a nocturnal predator that is known to feed on a wide variety of prey, such as fish, frogs, crabs, small mammals, birds, and reptiles. It is also reported to occasionally feed on carrion (Ali 2002). Published literature on the diet composition of *K. zeylonensis* in India is sparse. A study conducted by Vyas et al. (2013) on the breeding behaviour of *K. zeylonensis* in Jambughoda WS and surrounding areas in Gujarat, India reported fishes, crabs, insects, and prawns in the pellets of the species. However, the authors identified

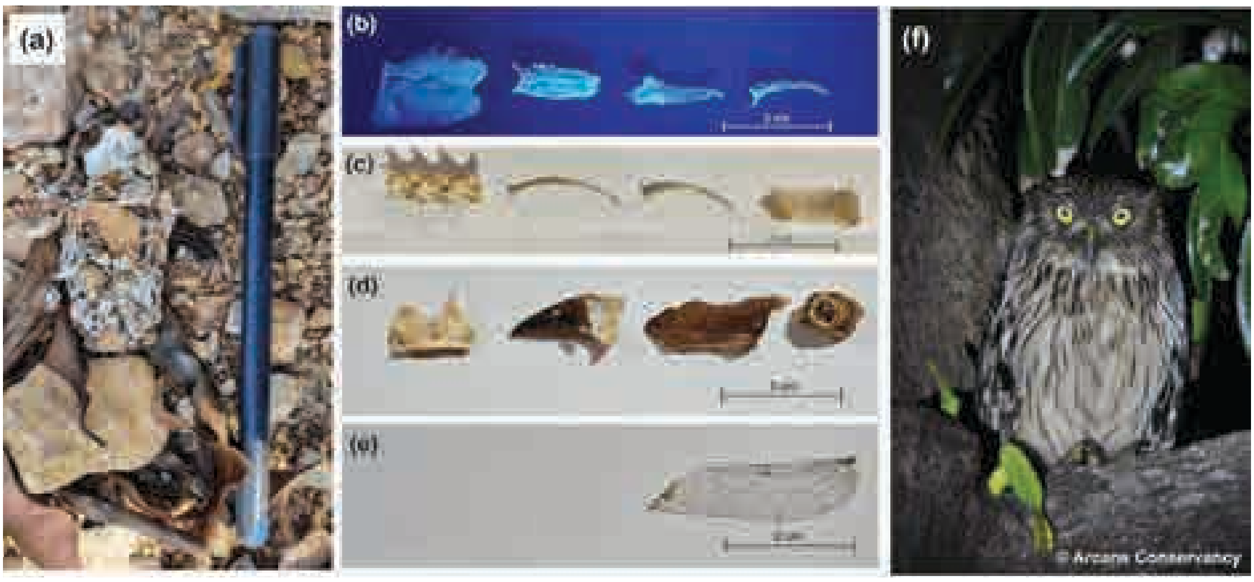

Image 1. Pellet analysis of Brown Fish-Owl *Ketupa zeylonensis*: a—A typical pellet deposited along a stream bank | b—Scorpion remains exhibiting fluorescence under ultraviolet light | c—Snake vertebral column, ribs, and skin | d—Crab remains | e—Insect remains (odonate wing) (© Stephen Jonah Dias & Atul Sinai Borker) | f—A Brown Fish-Owl on its perch in Study Area 2 (© Arcane Conservancy).

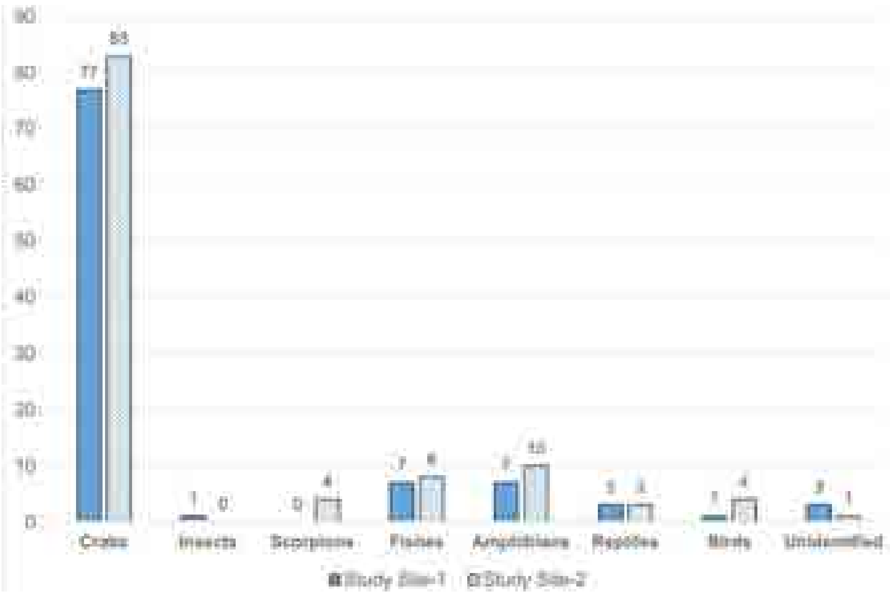

Figure 2. A comparison between prey categories recorded in the pellets of *Ketupa zeylonensis* in the study areas.

Table 1. Details of key body parts examined for the identification of the number of prey individuals in each pellet.

Prey Category	Key body parts used for assessing the number of individuals	Details of analysis
Crabs	Mouthparts, chelipeds, carapace, abdomen	The number of duplicates of exoskeletal structures (either whole parts or fragments) was used to estimate the number of individuals in each pellet.
Insects	Wings	
Scorpions	Pedipalps, cephalothorax shields, and telson	
Amphibians (Frogs)	Mouthparts, vertebrae (e.g., urostyle), pelvic girdle, humerus, radio-ulna, femur, tibio-fibula, and astragalus-calcaneum.	
Fishes	Parts of the axial skeleton (skull, vertebrae, and ribs), and scales.	Microscopic examinations of the morphological patterns on fish scales were conducted based on the principle that the patterns serve as useful taxonomic identifiers of fish species (Bräger & Moritz 2016). This was further supported by observations of the bones from the axial skeleton. As it was difficult to determine the number of individuals of the same species, all endoskeletal remains of similar size were assumed to be derived from a single individual unless morphological examinations of the scales indicated more than one species in the pellet.
Reptiles (Snakes)	Vertebrae, ribs, and skin	Identifying the number of individual snakes was straightforward in instances where the vertebral column was found to be relatively intact in the pellets. However, in instances where the vertebral column was found to be in a dismantled state, we used the general shape and size of the vertebrae and ribs to estimate the number of individuals. This was further supplemented by the remnants of snake skin present in the pellets.
Birds	Parts of the endoskeleton and feathers.	The number of duplicate endoskeletal remains was utilized to estimate the number of individuals. In cases where only feathers were present, feathers having similar morphological characteristics were assumed to originate from a single individual.

several other prey groups such as amphibians, reptiles, and birds from direct feeding observations and analysis of discarded prey items at the nests. This indicates that pellet analysis when supplemented with other observational protocols can significantly aid in the understanding of the food spectrum of the species. The diet composition of *K. zeylonensis* in Jambughoda WS was very similar to our observations in the Western Ghats of Goa with minor differences (Figure 3). In addition, the study in Jambughoda Wildlife Sanctuary was conducted during the pre-monsoon season (March–April) as compared to the present study that was conducted during the post-monsoon and winter seasons (October–February). Furthermore, fish owls are specialist birds of prey that have preferences for certain prey groups (Sieradzki 2023). Our data analysis supports this fact as the food niche breadth assessment indicated that *K. zeylonensis* is a specialist predator that feeds mainly on crabs whilst supplementing its diet with other invertebrate and vertebrate prey groups (Figure 2; Table 2).

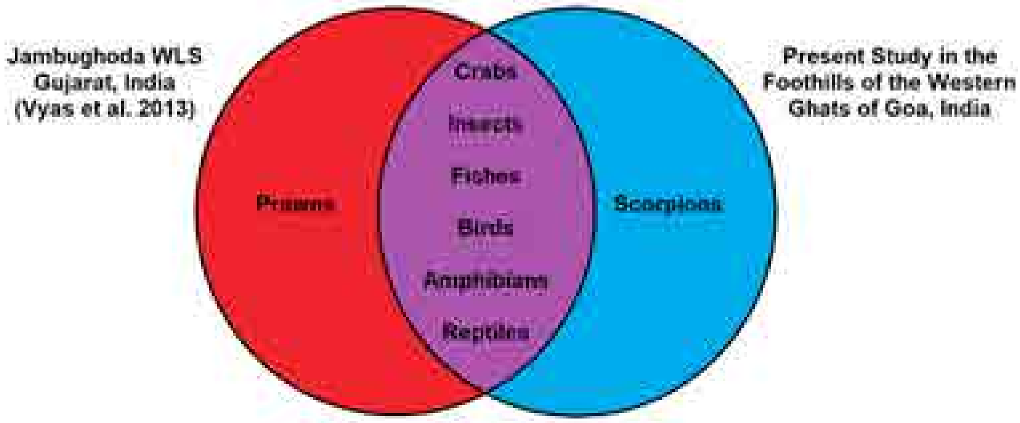

Figure 3. A comparative account of the diet of the Brown Fish-Owl *Ketupa zeylonensis* between Jambughoda Wildlife Sanctuary, Gujarat, and the present study conducted in the foothills of the Western Ghats of Goa, India

Table 2. Diet composition of the Brown Fish-Owl *Ketupa zeylonensis* in the foothills of the Western Ghats of Goa.

Phylum	Prey category	n	RFO %	FNB
Arthropoda	Crabs	160	75.47	0.1
	Insects	1	0.47	
	Scorpions	4	1.89	
Chordata	Fishes	15	7.08	
	Amphibians	17	8.02	
	Reptiles	6	2.83	
	Birds	5	2.36	
	Unidentified	4	1.89	
Total		**212**	**100**	

n—Number of individuals in each prey category | RFO %—Relative frequency of occurrence | FNB—Food niche breadth.

Pellet analysis is considered to be a robust indicator of the food spectrum of owls. In addition, such analysis can shed light on the richness, evenness, and abundance of prey groups constituting owl diet in the foraging environments (Heisler et al. 2015; Andrade et al. 2016). The present study was conducted due to the gap in knowledge in regards to the diet composition of *K. zeylonensis* in the Western Ghats ecoregion, particularly in the state of Goa. However, it is imperative to note that pellet collection in the present study was conducted for a relatively short period of time (post-monsoon and winter seasons), and the diet composition of owls is reported to change based on seasonal variations in prey availability (Kafkaletou-Diez et al. 2008; Santhanakrishnan et al. 2010). This may be an important factor to consider in landscapes such as the Western Ghats that undergo changes in hydrology across seasons. Organisms in such aquatic ecosystems may exhibit population changes on a seasonal scale that may influence the diet composition of the Brown Fish-Owl. Therefore, further assessments are required to understand the trends in the diet composition of the species across a seasonal gradient in the Western Ghats landscape.

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Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23521–23528
ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)
https://doi.org/10.11609/jott.8214.15.7.23521-23528
#8214 | Received 01 October 2022 | Final received 25 May 2023 | Finally accepted 06 July 2023

COMMUNICATION

Tree cover and built-up area regulate the territory size in Eurasian Magpie *Pica pica* in Ladakh, India

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Abstract: Eurasian Magpie *Pica pica* is one of the well-studied corvids, but the majority of our understanding of this species is from Europe. In India, its distribution is restricted to some valleys of Ladakh such as the northwestern part of the Indus, Nubra, Zanskar, Drass, and Suru. The present study aimed at understanding the territorial behavior of this species in small urban settlements of Ladakh region. Twenty-five pairs were studied in March 2020–April 2021. Territories were outlined for each color-banded individual, and data on habitat variables (namely built-up, agriculture, and green cover) was extracted. Generalized linear mixed models were used to study the effect of the habitat structure on territory size. The territory size (Mean ± SD) was 0.042 ± 0.025 km^2, with tree cover comprising the highest proportion (24.36 ± 15.41 %) of area within territories. Built-up area was a feature of all territories, highlighting the affinity of magpies towards human presence. Presence of tree cover and built-up area significantly (~ <0.002) reduced territory size. High adaptability, foraging, and nesting opportunities, and protection from predators have been recognized as the reasons for magpies' affinity with human habitation. Foraging opportunities are minimal outside human settlements in this region, magpies' territories are largely shaped by the fulfilment of foraging requirements.

Keywords: Behavior, Corvid, foraging, Himalaya, territorial, territory sharing, urban settlements.

Editor: Anonymity requested. **Date of publication:** 26 July 2023 (online & print)

Citation: Khan, I.A., A. Kumar, D. Bhatt & P. Rawal (2023). Tree cover and built-up area regulate the territory size in Eurasian Magpie *Pica pica* in Ladakh, India. *Journal of Threatened Taxa* 15(7): 23521–23528. https://doi.org/10.11609/jott.8214.15.7.23521-23528

Funding: None.

Competing interests: The authors declare no competing interests.

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Author contributions: IAK undertook field surveys, colour-banded the focal individuals of Eurasian Magpie, noted the behavioural data, took the images in the field and collected the literature for manuscript preparation. AK designed and planned the study including interpretation of data and manuscript writing. DB checked the manuscript and provided inputs for improvements. PR undertook the statistical analysis, prepared graphs and provided inputs for the interpretation of the statistical outcome.

Acknowledgements: Authors are grateful to Dr. Dhriti Banerjee, director, ZSI, Kolkata, for support and encouragements. First and second authors are thankful to officer-in-charge, ZSI, Dehradun, for institutional facilities. The first author is grateful to Prof. Dinesh Bhatt (Emeritus professor, Gurukul Kangri (Deemed to be University), Haridwar), Dr. Anil Kumar, scientist-E, ZSI, Dehradun, and Prof. D.S. Malik, head, Department of Zoology and Environmental Science, Gurukul Kangri, Haridwar, for departmental support to carry out this study. First author is thankful to Prakhar Rawal for his contribution in this study. First author is grateful to Ministry of Tribal Affairs (NFST) to grant the Ph.D. fellowship. Constant support and encouragements from, wildlife warden Kargil, and field staff of Wildlife Protection Division, Kargil is highly acknowledged.

UNIVERSITY OF TURKU

INTRODUCTION

Territory is an area defended by an organism or a group of organisms for mating, nesting, roosting, and foraging. During the breeding season, songbirds show territoriality in which the mated pairs defend the nest and feeding grounds until the young ones fledge (Alcock 2009). The size of the territory varies, depending on the habitat quality, structure, and the number of conspecific neighbors (Jones 2001; Flockhart et al. 2016; Skorupski et al. 2018). In urban areas, territory size differs in conspecific individuals depending on their ability to adapt to urban environments (Juarez et al. 2020). Territory size is crucial for breeding success which plays a major role for the survival and sustainability of species (Flockhart et al. 2016; Phillips et al. 2020). Hence, understanding of the territorial behavior is not only an interesting ecological inquiry but can also provide insights to manage landscapes, particularly urban ones, in a manner that can aid in the conservation of desired species.

The Eurasian Magpie *Pica pica* is a medium-sized corvid; an omnivorous bird with a range that includes Asia, Europe, and parts of northwestern Africa. Magpies often defend a vast, multi-purpose territory in which they nest, forage, and spend the majority of their time (Birkhead 1991). Eurasian Magpies are an urban adapter species, capable of invading towns while also maintaining a wild population in rural and natural habitats (Jokimäki 2017). Although it is one of the most studied species of corvids with majority of the research conducted in Europe. Studies on the bird in other continents are still scanty (Benmazouz et al. 2021). Magpies have a high level of fidelity to their home range, indicating that their dispersal lengths are quite small (Birkhead 1991).

Ladakh is characterized by large stretches of uninhabited land interspersed with small human settlements where magpies can be found. Magpies are known to be sedentary and usually do not migrate among these villages, and they act as isolated habitats rather than a gradient, with no individuals observed in between (Newton 2010). The study of bird territorial behavior in such isolated systems can help us understand how territorial individuals coexist in small habitats. Studies on magpies from these high-altitude regions of Ladakh are virtually absent (Khan et al. 2022).

In this study, we investigated the territorial behavior of Eurasian Magpies in the small, isolated urban settlements of Ladakh. Our preliminary findings revealed that the distribution of the species in Ladakh is patchy, with most populations confined to areas with human settlements. We assumed that human settlements might have an impact on the daily activities of magpies, either directly or indirectly. According to previous research, magpies are more attracted to man-made food scraps, which reduces magpie hunting and natural food consumption (Croci et al. 2008; Jokimaki et al. 2017; Salek et al. 2020). Based on this, we predicted that (1) magpie territory would be smaller near built-up areas due to increased food provisioning and (2) territory with a higher proportion of tree cover would be smaller in size because tree patches provide all essential food resources. We also predicted that (3) an open area with fewer tree patches would have lower food production and that the magpie's territory size would be larger in order to meet the food requirements.

MATERIALS AND METHODS

Study area

The research was carried out at two locations, namely Gramthang village of Suru Valley and Bursaika village of Wakha Valley in the district of Kargil (Image 2), Ladakh of India. About 8 km^2 area at each location was explored for study. Gramthang (34.467^{0}N, 76.084^{0}E) is situated about 12 km from Kargil. It is located at an average elevation of 2750 m and has river-fed well-vegetated lands with a high concentration of *Populus alba*, *P. ciliata*, *P. nigra*, *Prunus armeniaca*, *Salix alba*, *S. excelsa*, and *S. fragilis* plantations. Bursaika (34.366^{0}N, 76.383^{0}E) is 40 km from Kargil and is part of the Wakha Valley with an elevation of 3,450 m. The landscape consists of open, arable cropland, patchy shrublands, a moist meadow with perennial spring water, and *Salix* vegetation. The number of *Populus* trees plantation in Bursaika are substantially smaller than in Gramthang due to water constraints and harsh terrain. Instead, the vegetation is comprised of *Salix fragilis* and Sea buckthorn *Hippophae rhamnoides* shrubs, with fewer *P. alba*. The summer temperature in Gramthang ranges from 10°–25° C, while the winter temperature can reach -29° C at its coldest. Bursaika winters are colder, with temperatures dropping to -35° C during peak winters (Khan et al. 2022).

Behavioral observations and territory marking

Twenty-five breeding individuals were caught using bait traps. The method was adopted from a past study (Kautz & Seamans 1992) and color-banded for individual identification (Image 2). In 2019 and 2020, the same individuals were seen at the sites, indicating little to no migration. Territorial observations were made in the

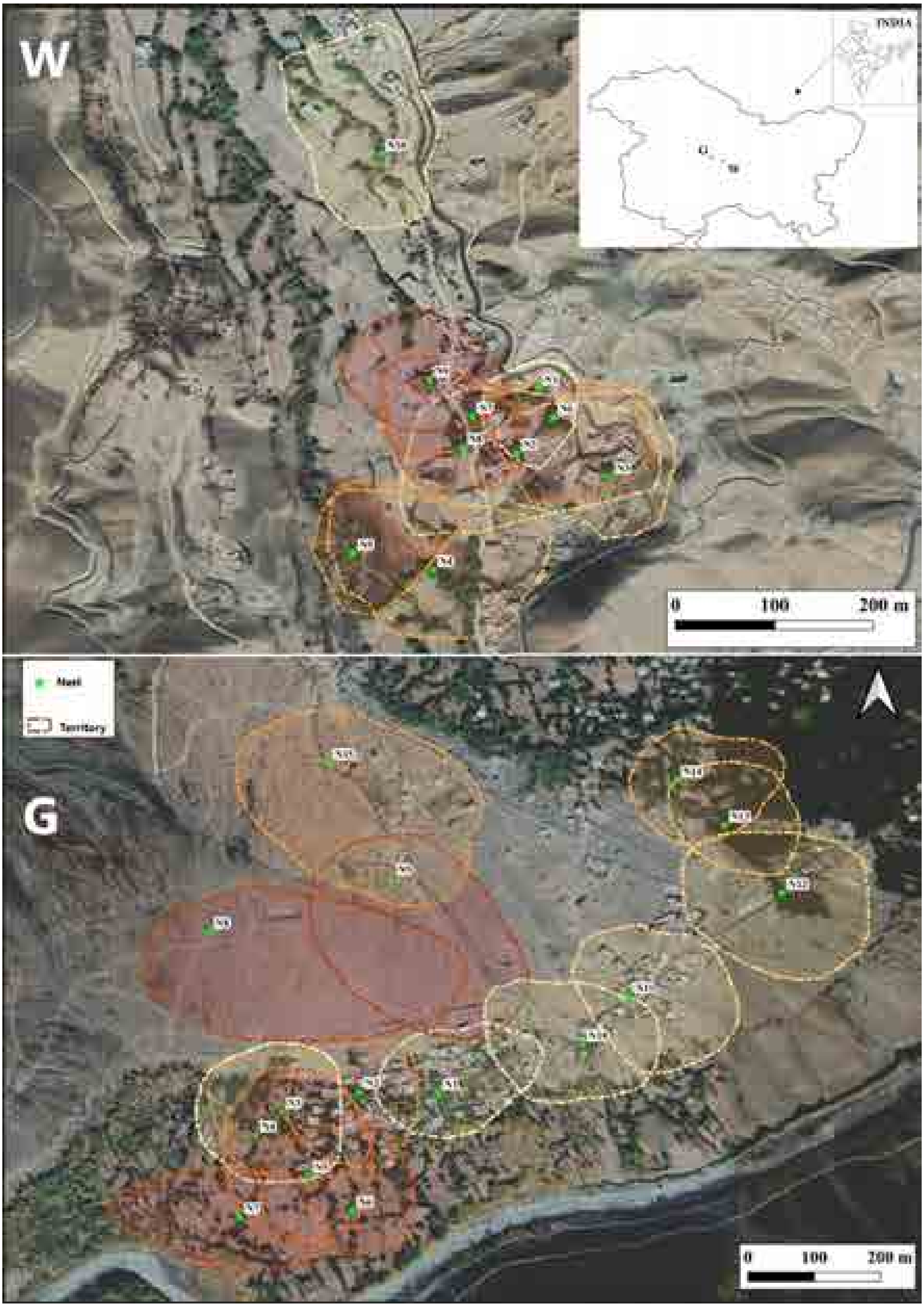

Image 1. Satellite imagery of Bursaika village in Wakha (above) and Gramthang village (G, below). The territories of individuals (assigned with a number) are marked with nest locations (N) and dotted boundaries. A gradient of color is used to differentiate territories. Small box in top right shows the location of two sites in India.

Image 2. The colour-banded tagged Eurasian Magpie *Pica pica* during nest/ territory vigilance: A—Colour banding of an adult individual | B—Adult individual released after banding | C—Colour-banded nestling | D—Another colour-banded nestling. © Iqbal Ali Khan.

months of March and April when the birds were nesting. Observations were carried out in 2020 and 2021 at sites Bursaika and Gramthang, respectively. Behavioral observations were made with field binoculars or with the naked eyes, depending on the situation. Nesting locations of territory owners were also discovered prior to egg laying by simply looking for birds carrying nest material. This was relatively easy in the early part of the season before the trees went into leaf in the summer (May–June). Since magpies are diurnal, each focal individual was tracked from a safe distance (about 10–30 m) for almost the whole day from early morning emergence time (0600–0630 h) to late roosting time (1830–1900 h). The locations visited by magpies for foraging, roosting, water drinking, and playing (Image 3, 4) were all tracked and marked using GPS (Garmin Etrex 30) shortly after the bird left the spot.

Variable extractions

Territorial variables included territory size, number of foraging points & the amount of tree cover, cultivated area, built-up area, and miscellaneous area (shrubland,

Image 3. A pair of Eurasian Magpie *Pica pica.* The male is seen here producing territorial calls. © Iqbal Ali Khan.

Image 4. The nesting female defending the nest. © Iqbal Ali Khan.

rock terrain, river stream, and grassy meadow) within the territory were extracted using polygons in Google Earth Pro software (version 7.3.6.9345). We determined the total area of the territory by connecting all the GPS points used by the focal pair of magpies during the breeding season, plotting all the points in the Google Earth satellite imagery and tracing out the total territory of the magpie by connecting all the points and forming a polygon. Other variables within the territory, such as tree patches, cultivated area, built-up area, and other miscellaneous areas were also traced using polygons. Multiple polygons were traced in one territory, and then all the polygons were combined to identify the different variable areas. Field notes and Google satellite images were used to cross-check all the sites and areas, and a high-resolution territories map was created. We studied the influence of neighbors by extracting the proportion of their territory which overlapped with the territory of other individuals.

Data analysis

The analysis was carried out using R version 4.2.2. As the territory size was not normally distributed (Shapiro-Wilk normality test, $p = 0.01$), and individuals were selected from two different sites, we used Generalized Linear Mixed Models (GLMM) to study the influence of the proportion of different land cover type on the territory size using the package lmerTest (Kuznetsova et al. 2017). Based on a correlation matrix, we removed the highly correlated ($r > |0.4|$) variables and selected 4 variables for the analysis – tree, agricultural, built-up cover, and neighbor presence. Their proportions were used, rather than the absolute area. The response variable was territory size in m^2, but the results are presented in km^2 for clarity. Sites (Gramthang & Bursaika) were taken as the random effects. We ran multiple models using different families and selected the best model based on AIC values. Regression plots were created using model results with the help of package effects (Fox & Weisberg 2019).

RESULTS

Descriptions of territories

We collected territorial data of 25 breeding pairs of Eurasian Magpies and observations showed that the magpie territory is almost circular in shape, with the nest being located close to the center. The breeding territory size of magpies varies from a minimum of 0.0094 km^2 to a maximum of 0.1049 km^2 (mean: 0.0415 ± 0.0248 km^2, n = 25) for all territories in the two sites. Magpie territories overlapped heavily, seen at both study sites, and magpies actively defended only the close proximity of the nesting tree (~ <20 m radius). Juveniles and non-breeding individuals (floaters) were occasionally spotted foraging in groups inside breeding territories of nesting pairs. Tree cover composed the highest amount of territory cover (mean proportion of territory for all individuals: 24.36 ± 15.41 %), followed by agricultural land (22.32 ± 15.51 %), and built-up areas (14.12 ± 9.73 %). All magpie territories in both sites feature human presence (mean proportion of territory for all individuals: 36.4 ± 19.13 %), either in the form of agricultural land or built-up areas, or both. Magpie territories in Bursaika were smaller (mean: 0.0212 ± 0.0084 km^2) and showed greater overlapping, with seven of the 10 individuals sharing more than 75 % of their territories (mean territory shared: 73.3 ± 30.5 %). The distance between nests at this site was also smaller, with an average distance of 81 m to the nearest nest. On the other hand, territories at Gramthang were larger (0.055 ± 0.0219 km^2) and with relatively lower territory sharing (55.6 ± 28.5 %). The average distance to the nearest nest was also larger at this site (134 m). The majority of the nests were located on *Populus* (9.22 ± 1.64 m; n = 9) and willow trees (6.62 ± 0.74 m; n = 8), followed by apricot (6.75 ± 0.95 m; n = 4), mulberry (8.5 ± 0.7 m; n = 2), and sea-buckthorn shrub (3.00 m; n = 1). Only one of the 25 nests was found on an artificial structure, an electric tower (in Gramthang). Nearly all nests (except a single nest on sea-buckthorn shrub), were constructed at a height >5 m.

Effect of habitat variables on territory size

We found that both increased built-up area and tree cover proportions within the territory had a significantly strong negative effect on the territory size of magpies, meaning that magpie territories are smaller near urban areas and greater tree cover (Figure 1). This is likely due to the high availability of resources near trees and urban areas, removing the need to defend large territories. Agriculture area had no significant effect, indicating limited feeding opportunities in agricultural fields during the study period. The presence of neighbors is also found to not have any significant effect, which is in-line with previous studies which have shown magpies to share feeding grounds. Table 1 summarizes the GLMM results describing the individual contributions of habitat variables in predicting territory size.

DISCUSSION

The current study describes the territorial behavior of Eurasian Magpies, and how territory size varies with habitat variables in the sparse urban settlements of the Himalayas. Characteristics of magpie territories, including choice of nesting sites, territory size, and territory sharing behavior, are largely similar to those observed in previous studies from other parts of the world. Previous studies have found magpie territory sizes to be 5 ha on average (Moller 1982; Birkhead 1991), but the mean can range anywhere from 1 ha–7.5 ha (Reese & Kadlec 1985; Dhindsa & Boag 1991). The mean territory size in our sites also lies within the expected range, with a mean of 4.15 ha. Although, only part of the territory close to the nest (~ within a 20 m radius of the nest) is actively defended by the breeding pair, other individuals entering this space aggressively pushed away. Magpie territories appear to be less rigidly defined, as both breeders and non-breeders can be found in the same spaces on subsequent visits. During breeding seasons, magpies were frequently seen chasing each other and calling from prominent perches with aggressive wing-fluttering. Although magpies are primarily territorial during breeding seasons, they are known to flock for 'ceremonial gatherings' (Baeyens 1979), roosting (Moller 1985), and feeding (Vogrin 1998). Magpies in our sites shared territories primary for feeding, gathering to feed at a few selected points where food waste was dumped. Magpies are likely to feed together, even during the breeding season, most probably owing to the limited food resources in this landscape, largely restricted to these small urban

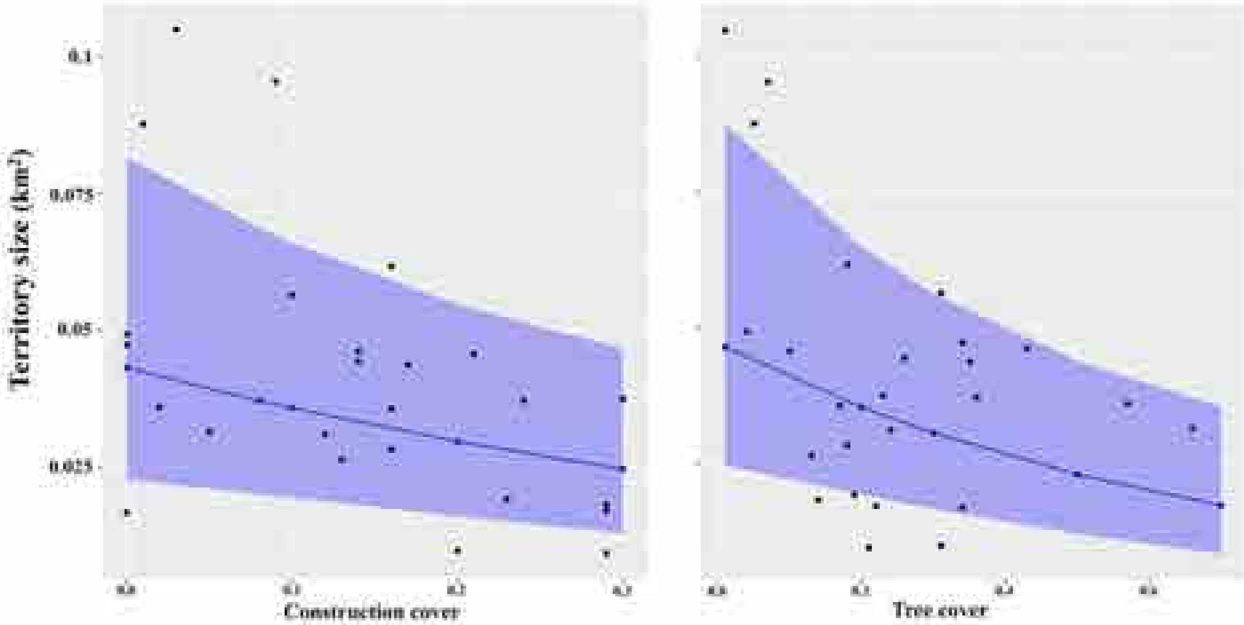

Figure 1. Linear response of territory size to construction and tree cover as obtained from results of generalized linear mixed model (GLMM).

settlements. This claim is further strengthened by the fact that magpies formed smaller territories and stayed closer to neighbors at the site, i.e., Wakha, pointing to the need for magpies to stay close to human habitation, even at the cost of sharing feeding spaces. Magpies have previously been observed to form feeding flocks when the resources are localized and clumped (Eden 1987).

Although magpies are widely known to be able to nest on artificial structures (Birkhead 1991; Takeishi 1994), they prefer to nest on trees, and only choose artificial nesting sites in case the tree density is low (Nakahara 2015). Additionally, in human habitations magpies construct their nests at greater heights (usually over 5 m), primarily to avoid human disturbance and predation (Antonov 2002, 2003; Salek 2020). Both *Populus* and willow trees, which were majorly used for nesting in the region, are tall trees providing suitable nesting sites for magpies (growing up to ~30 m) and have previously been shown to be preferred tree species for nesting of magpies (Antonov 2002). Moreover, large artificial structures are absent in the sparse urban settlements of this region, limiting opportunities for nesting on artificial structures. Therefore, all (except one) nests were constructed on trees. The sole nest constructed on an electric tower was away from housing, with no trees in close proximity.

Trees are not only an absolute necessity for nesting in these sites, but they may also be provisioning important food resources, like insects, butterfly/moth larvae at these sites. The other primary food source in magpie territories was human-dumped waste sites, as explained earlier. Urban adapter species are known to form smaller territories near human habitation due to high availability of resources in close proximity, such as waste dump (Juarez et al. 2020). Hence, in line with our predictions, we found the presence of both tree cover and built-up area to have a significant negative effect on territory size (Table 1). Additionally, due to the localization of resources to these small sites, magpies are willing to share feeding sites even during the breeding season. Therefore, in these sites the presence of neighboring magpies does not significantly affect territory size, indicating that the major driver of territory size in these isolated urban settlements is resource availability, rather than interspecific interactions. Tatner (1982) previously found no association between magpie

Table 1. Summary of GLMM results with values of coefficients, standard errors (SE) and p-value for the selected variables.

Variables	Coefficient	SE	P value
Intercept estimate	11.15	0.33	<0.001
Tree cover	- 1.44	0.41	<0.001
Built-up	- 1.87	0.62	0.002
Agriculture	- 0.41	0.36	0.26
Neighbor	-0.05	0.19	0.79

density and breeding success in urban areas, as long as the territory is resourceful. Magpies have previously been shown to prefer urban areas with suitable nesting sites and trees from different parts of the world (Wang et al. 2008; Salek et al. 2020), and we add to the existing knowledge from the isolated urban settlements of the Himalaya, for the first time.

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Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23529–23544

ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)

https://doi.org/10.11609/jott.7520.15.7.23529-23544

#7520 | Received 13 June 2021 | Final received 19 June 2023 | Finally accepted 30 June 2023

OPEN ACCESS

COMMUNICATION

Birds of Kanetiya area - inventory, notable sightings, and overview of seasonal changes in reporting frequency of bird species in an unprotected area of Himachal Pradesh, India

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Abstract: Biodiversity of unprotected areas in the western Himalayan region is under threat. Despite this, it is poorly studied and documented. The citizen science platform eBird was used to record bird species of the unprotected Kanetiya area (Darbhog panchayat), Shimla, Himachal Pradesh from August 2019–2020. Reporting frequency using this data was calculated to represent an index of species abundance. This was calculated independently for each species across three seasons and reported as a metric that can be tracked over time. One-hundred-and-twenty-four (20% of the species from Himachal Pradesh) species of birds belonging to 13 orders and 43 families were recorded. Of these, 37 (30%) were recorded year-round and the remaining 87% (80%) were migratory. The checklist consisted of five species of high conservation concern and 22 species of moderate conservation concern. This checklist also provides insights into the distributions of some species whose ranges within India are not yet well defined (Northern Long-eared Owl *Asio otus*, Aberrant Bush Warbler *Horornis flavolivaceus*, Himalayan Owl *Strix nivicolum*) and into migration through this part of the Himalaya (Black Stork *Ciconia nigra*). Locals can be educated to upload short checklists for monitoring since they have helped the local forest department's conservation efforts.

Keywords: Abundance, checklist, eBird, reporting frequency, western Himalaya.

Editor: Rajiv S. Kalsi, M.L.N. College, Haryana, India. **Date of publication:** 26 July 2023 (online & print)

Citation: Tiwari, S. (2023). Birds of Kanetiya area - inventory, notable sightings, and overview of seasonal changes in reporting frequency of bird species in an unprotected area of Himachal Pradesh, India. *Journal of Threatened Taxa* 15(7): 23529–23544. https://doi.org/10.11609/jott.7520.15.7.23529-23544

Funding: None.

Competing interests: The author declares no competing interests.

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Acknowledgments: I thank Ashwin Viswanathan for commenting on various drafts of the manuscript and suggesting the method for analyzing the data and reporting frequency. I also thank Sandhya Marathe and Monica Kaushik for commenting on early stages of the manuscript. I am grateful to residents, especially, Vikas Thakur, Sanjeev Kumar, Satish Thakur, Laikram Thakur, Mamta Thakur, and Dhiman for sharing their personal observations with me. I am grateful to Puja Sharma, Tim Inskipp, Rajah Jayapal, and staff of SACON and WII library for providing various reference materials used in the manuscript. I also thank the reviewers and the subject editor for their valuable suggestions and constructive criticism. For methodology, I also referred to https://www.slideshare.net/suhelq/neighbourhood-bird-monitoring-through-consistent-listing.

INTRODUCTION

Himachal Pradesh in the western Himalaya is home to several species of animals and plants. The protected area network of the state consists of five national parks, 26 wildlife sanctuaries, and three conservation reserves spread across 8,391 km^2 (Himachal Pradesh Forest Department 2022). Apart from this, several species are found in the unprotected region, which makes up more than 85% of the state. These include several endemic and threatened species like the Musk Deer *Moschus moschiferus*, Cheer Pheasant *Catreus wallichii*, and Himalayan Yew *Taxus wallichiana*. Although unprotected areas of the western Himalaya support biodiversity, they are threatened by deforestation, habitat alteration and habitat fragmentation caused by the construction of roads and trails (Pandit et al. 2007; Pandit & Kumar 2013). Habitats of such areas may change or degrade completely, leading to extinctions even before the documentation of their biodiversity is complete (González-Oreja 2008). Conversion of natural habitat can specifically lead to local extinctions of specialist species across various taxa (Korkeamäki & Suhonen 2002; Munday 2004). For example, the Vulnerable Cheer Pheasant became locally extinct in Jaunaji, Himachal Pradesh, after grasslands were converted into agricultural lands (Kaul 2014). Medicinal plants like the Elephant's Foot *Dioscorea deltoidea* and Himalayan Yew are threatened with extinction due to overexploitation (IUCN 2008). In private landholdings of rural areas, local communities often burn grasses and understories to increase the yield of grass in summer (Garson et al. 1992). This endangers native ground-dwelling birds and other fauna (Manupriya 2019).

Due to these concerns, scientists, conservation managers, and local communities must focus on monitoring and devising ways to conserve these habitats (Herremans 1998) and the species they support. This will require an inventory of taxa found in different regions (Llanos et al. 2011; Sharma et al. 2018) and an understanding of the effects of land use change on various floral and faunal communities.

Birds can be used as model taxa to understand the biodiversity health of an ecosystem (Eglington et al. 2012). This is because they play diverse roles in an ecosystem (e.g., pollinators, seed dispersers) (Garcia et al. 2010; Whelan et al. 2015) and have an intricate association with their environment. Subsequently, areas that support many birds of high conservation concern can be prioritized for conservation. Repeated surveys can also draw attention to the decline in functional diversity of bird species from an area. This can further highlight the degradation in ecosystem services like decomposition, pollination, and seed dispersal (Şekercioğlu et al. 2004).

An informative baseline checklist of the birds of the human-dominated Kanetiya area in Shimla, Himachal Pradesh is presented in this study. This landscape lies in Darbhog panchayat, Shimla Rural tehsil. It lies outside the protected area network and is shaped by various anthropogenic activities of the residents. Reporting frequency has been used to provide an index of the seasonal abundance of each species. This can be used as a baseline to assess the change in species composition with time.

MATERIALS AND METHODS

Study area

This study uses checklists submitted by the author [ST] while visiting Seri, Bagdra, Jalpan, Kool, and Undala villages and their surroundings. These villages lie within the Kanetiya region (Figure 1) named after the local deity Kanetiya Maharaj. The region comes under the jurisdiction of the Darbhog panchayat, Shimla rural tehsil.

The surveyed area spreads across 3.5 km^2. Its elevation ranges from c. 1,480–2,190 m, between 31.0340–31.0115 °N and 77.2764–77.3004 °E. A tributary of the river Yamuna flows through the lowest part of the sampled area. The landscape is highly fragmented and comprises plant communities either dominated by Banj Oak *Quercus leucotrichophora*, Deodar *Cedrus deodara*, or grasslands scattered with Chir Pine *Pinus roxburghii*. The area has a temperate climate and the temperature ranges from -9–31 °C. Snowfall occurs in the area almost every year, and in January 2020, it reached an eight-year high (Press Trust of India 2020). Residents used the area for fodder collection, resin, wood collection, cattle grazing and religious purposes. In June 2019, a forest patch of the area suffered from a fire that had spread to it from nearby grasslands.

The area lies 22 km from Chail Wildlife Sanctuary, 26 km from Churdhar Wildlife Sanctuary, and 8 km from Shimla Water Catchment Wildlife Sanctuary (Google Earth Pro 2020). Though it lies outside sanctuaries and national parks, the Himachal Pradesh Forest Department in cooperation from residents has reintroduced the Vulnerable Cheer Pheasant in grasslands between Seri and Undala villages (IUCN 2020b). The reintroduction site consists of a demarcated intensive management area that spreads across one square kilometer and consists of grasslands and demarcated protected forests.

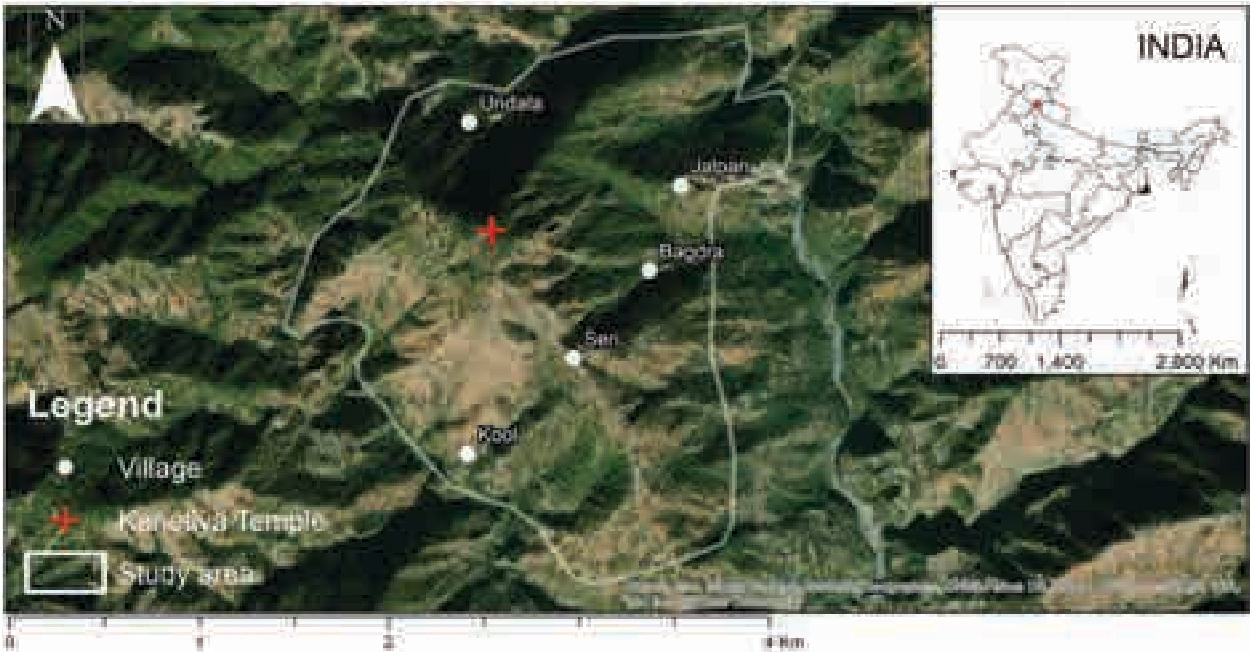

Figure 1. The region within the Kanetiya area, Shimla where birds were recorded from August 2019–2020.

Data collection

The bird checklists were uploaded using the eBird mobile app while visiting the region between August 2019 and August 2020. When all identified species were reported, the checklists were deemed complete; however, if some species were deliberately omitted, they were considered incomplete. Along with these details, the checklists included the date, starting time, duration, observation type (stationary/traveling) and track record. Species were recognized visually or by call, and checked with field guides (Grimmett et al. 2011; Rasmussen & Anderton 2012) and the Merlin picture identification app (The Cornell Laboratory 2020) for confirmation. Photographs and call recordings of unidentified species were shared with experts for identification.

Around 341 complete checklists from the eBird website were downloaded and combined with 11 complete checklists uploaded by other eBird users who visited the area during the study period. If a checklist had been shared with multiple observers, the version with the maximum number of species was chosen. In addition to the eBird collection, a local reported one species (Cattle Egret *Bulcus ibis*). A total of 212 checklists (60%) were less than an hour long, 107 checklists ranged in length from one to two hours and 24 checklists, each lasting between two and three hours. Two checklists were 4 to 5 hours long, while seven checklists ranged in length 3–4 hours. The checklists concerned the three seasons. These were the summer season (April through June; effort: 80 checklists), the monsoon season (July through September; effort: 37 checklists), and the winter season (October through March; effort: 235 checklists). The dataset included 100 stationary checklists (summer 22, monsoon 7, and winter 71) and 252 traveling checklists (summer 58, monsoon 29, and winter 165). Throughout the course of the research, 346.28 hours were put in (summer: 76.13; monsoon: 27.12; winter: 243.03), and 129.09 km were traveled (summer: 23.6; monsoon: 22.96; winter: 82.53).

Data analysis

Microsoft Excel 2007 was used to organize the data and calculate the reporting frequency of each species across different seasons. Reporting frequency is the percentage of checklists in which a species was recorded over a given period ((number of checklists a species was recorded during a season/number of total complete checklists reported during the season) X 100) (Viswanathan et al. 2020; eBird 2021c). Reporting frequency was calculated for each species separately for three seasons.

Species were classified as 'year-round' if they were reported across all three seasons, and 'seasonal' if they were detected only during certain seasons. India checklist v4.0 (Praveen et al. 2020) and IUCN Red List (IUCN 2020a) were used to refer to the taxonomy of species and their threat status, respectively. State of India's Birds Report (SoIB 2020a) was used to categorize birds as per their status of conservation concern. This report used short-term and long-term population trends of species to categorize them as species of high, moderate, and low conservation concern.

RESULTS

One-hundred-and -twenty-four bird species belonging to 13 orders and 43 families were recorded. Of these, 37 were present year-round and 87 were seasonal. 74, 57, and 101 species were recorded in summer, monsoon, and winter, respectively.

Five species of high conservation concern (SoIB 2020a) were recorded during the study. All five had a low reporting frequency. These were Cheer Pheasant *Catreus wallichii* (summer-10, winter- 2.55), Red-headed Vulture *Sarcogyps calvus* (winter- 1.70), Short-toed Snake Eagle *Circaetus gallicus* (summer-1.25, winter- 6.38), Steppe Eagle *Aquila nipalensis* (monsoon-2.70, winter- 11.06), and White-rumped Vulture *Gyps bengalensis* (winter- 0.85). None of these were record in all three seasons.

About 22 species of moderate conservation concern were identified (SoIB 2020a). In all three seasons (summer, monsoon, and winter), the Himalayan Griffon *Gyps himalayensis* had the highest reported frequency. In the summer and monsoon seasons, the Upland Pipit *Anthus sylvanus* was seen to report a frequency of 52.5 and 32.43, respectively. Other species of moderate conservation concern included the Grey-headed Canary-flycatcher *Culicicapa ceylonensis*, Long-tailed Minivet *Pericrocotus ethologus*, Lemon-rumped Warbler *Phylloscopus chloronotus*, and Common Kestrel *Falco tinnunculus*, all of which had very low reporting frequencies in all three seasons (<10) (Table 1).

Of the remaining 97 species, 89 were of low conservation concern and eight had not been categorized.

The most frequently observed species during the summer were Striated Prinia *Prinia crinigera* (68.75), Himalayan Bulbul *Pycnonotus leucogenis* (67.5), Great Barbet *Psilopogon virens* (66.25), Large-billed Crow *Corvus macrorhynchos* (65), Blue-throated Barbet *Psilopogon asiaticus* (52.5) and Upland Pipit *Anthus sylvanus* (52.5) (Figure 2). Of these, the Blue-throated Barbet *Psilopogon asiaticus* and Upland Pipit *Anthus sylvanus* were designated as seasonal. In the monsoon season, Himalayan Bulbul *Pycnonotus leucogenis* (78.38), Great Barbet *Psilopogon virens* (43.24), Striated Prinia *Prinia crinigera* (40.54), Large-billed Crow *Corvus macrorhynchos* (40.54), Black Francolin *Francolinus francolinus* (40.54) and Upland Pipit *Anthus sylvanus* (32.43) were reported most frequently. All of the species that were most frequently reported during the winter were recorded all year round. These include the Large-billed Crow *Corvus macrorhynchos* (71.06), Himalayan Bulbul *Pycnonotus leucogenis* (61.28), Himalayan Griffon *Gyps himalayensis* (50.64), Blue Whistling Thrush *Myophonus caeruleus* (33.19), Grey-hooded Warbler *Phylloscopus xanthoschistos* (29.36), and Great Barbet *Psilopogon virens* (27.23).

Thirty-seven species were recorded in all three seasons and classified as year-round or resident. These included species of moderate conservation concern like the Himalayan Griffon *Gyps himalayensis* (summer- 37.5, monsoon-27.03, winter- 50.64), Common Kestrel *Falco tinnunculus* (summer- 6.25, monsoon- 5.41, winter- 5.53), Long-tailed Minivet *Pericrocotus ethologus* (summer- 2.5, monsoon- 2.70, winter- 2.13), Lemon-rumped Warbler *Phylloscopus chloronotus* (summer- 2.5, monsoon- 2.70, winter- 2.98), and Grey-headed Canary-flycatcher *Culicicapa ceylonensis* (summer- 1.25, monsoon- 2.70, winter- 0.43).

Of the year-round species, 17 had the highest reporting frequency during summer (Figure 2A), 12 during the monsoon (Figure 2B) and eight during the winter season (Figure 2C).

Thirteen species were exclusively recorded during the summer. Three of these, Black Stork *Ciconia nigra* (an incidental record), Plumbeous Water Redstart *Phoenicurus fuliginosus* (an incidental record) and Himalayan Cuckoo *Cuculus saturates* (11.25) were of moderate conservation concern. Of the species recorded exclusively during the monsoon, three, namely Asian Brown Flycatcher *Muscicapa dauurica* (2.7) and Black Redstart *Phoenicurus ochruros* (5.41) were of moderate conservation concern and the Chestnut-bellied Rock Thrush *Monticola rufiventris* (2.70) was of low conservation concern 37 species were recorded exclusively during the winter. Among these, raptors like the Red-headed Vulture *Sarcogyps calvus* (1.70) and White-rumped Vulture *Gyps bengalensis* (0.85) were of high conservation concern. A few species of moderate conservation concern like Koklass Pheasant *Pucrasia macrosomia* (0.43), Golden Eagle *Aquila chrysaetos* (0.85), Long-tailed Shrike *Lanius schach* (1.70), Altai Accentor *Prunella himalayana* (0.43), Black-throated Accentor *Prunella atrogularis* (2.13), Himalayan White-browed Rosefinch *Carpodacus thura* (0.43), and White-capped Bunting *Emberiza stewarti* (0.43) were exclusively recorded during this season.

Significant sightings

The following records are significant as they provide

Table 1. Checklist of bird species recorded in the Kanetiya region (3.5 km²) from August 2019–2020 along with the IUCN category (IUCN 2020a), category of conservation concern (SoIB 2020a) and reporting frequency across seasons.

	Common name	Scientific name	IUCN Red List status	Status of conservation concern	Summer (April–June) (80 checklists)	Monsoon (July–September) (36 checklists)	Winter (October–March) (235 checklists)	Migratory status
1	Indian Peafowl	*Pavo cristatus*	LC	L	3.75	0	1.28	S
2	Black Francolin	*Francolinus francolinus*	LC	L	47.5	40.54	15.74	YR
3	Red Junglefowl	*Gallus gallus*	LC	L	7.5	2.70	0	S
4	Cheer Pheasant	*Catreus wallichii*	VU	H	10	0	2.55	S
5	Kalij Pheasant	*Lophura leucomelanos*	LC	L	27.5	13.51	12.34	YR
6	Koklass Pheasant	*Pucrasia macrolopha*	LC	M	0	0	0.43	S
7	Rock Pigeon	*Columba livia*	LC	L	2.5	2.70	0.85	YR
8	Oriental Turtle Dove	*Streptopelia orientalis*	LC	L	17.5	10.81	0.85	YR
9	Wedge-tailed Green Pigeon	*Treron sphenurus*	LC	L	6.25	0	0	S
10	Himalayan Cuckoo	*Cuculus saturatus*	LC	M	11.25	0	0	S
11	Common Cuckoo	*Cuculus canorus*	LC	M	35	2.70	0	S
12	Grey Nightjar	*Caprimulgus jotaka*	LC	ND	15	0	0	S
13	Black Stork	*Ciconia nigra*	LC	M	I*	0	0	S
14	Cattle Egret	*Bubulcus ibis*	LC	L	0	0	I*	S
15	Bearded Vulture	*Gypaetus barbatus*	NT	M	1.25	0	2.55	S
16	Oriental Honey Buzzard	*Pernis ptilorhynchus*	LC	L	I*	0	0	S
17	Red-headed Vulture	*Sarcogyps calvus*	CR	H	0	0	1.70	S
18	White-rumped Vulture	*Gyps bengalensis*	CR	H	0	0	0.85	S
19	Himalayan Griffon	*Gyps himalayensis*	NT	M	37.5	27.03	50.64	YR
20	Short-toed Snake Eagle	*Circaetus gallicus*	LC	H	1.25	0	6.38	S
21	Mountain Hawk Eagle	*Nisaetus nipalensis*	LC	L	I*	0	I*	S
22	Booted Eagle	*Hieraaetus pennatus*	LC	L	1.25	0	0	S
23	Steppe Eagle	*Aquila nipalensis*	EN	H	0	2.70	11.06	S
24	Golden Eagle	*Aquila chrysaetos*	LC	M	0	0	0.85	S
25	Bonelli's Eagle	*Aquila fasciata*	LC	L	1.25	0	2.98	S
26	Hen Harrier	*Circus cyaneus*	LC	ND	3.75	0	2.55	S
27	Shikra	*Accipiter badius*	LC	L	0	2.70	2.13	S
28	Mountain Scops Owl	*Otus spilocephalus*	LC	ND	6.25	0	4.68	S
29	Collared Owlet	*Glaucidium brodiei*	LC	L	0	0	0.43	S
30	Asian Barred Owlet	*Glaucidium cuculoides*	LC	L	0	0	2.98	S
31	Himalayan Owl	*Strix nivicolum*	LC	ND	I*	0	0	S
32	Northern Long-eared Owl	*Asio otus*	LC	ND	0	0	2.55	S
33	Common Hoopoe	*Upupa epops*	LC	M	3.75	2.70	0	S
34	Great Barbet	*Psilopogon virens*	LC	L	66.25	43.24	27.23	YR
35	Blue-throated Barbet	*Psilopogon asiaticus*	LC	L	52.5	13.51	0	S
36	Speckled Piculet	*Picumnus innominatus*	LC	L	0	0	1.28	S
37	Brown-fronted Woodpecker	*Dendrocoptes auriceps*	LC	L	2.5	2.70	9.36	YR
38	Fulvous-breasted Woodpecker	*Dendrocopos macei*	LC	L	0	2.70	2.13	S
39	Himalayan Woodpecker	*Dendrocopos himalayensis*	LC	L	0	2.70	2.13	S
40	Lesser Yellownape	*Picus chlorolophus*	LC	L	2.5	0	0	S
41	Scaly-bellied Woodpecker	*Picus squamatus*	LC	L	28.75	27.03	19.15	YR
42	Grey-headed Woodpecker	*Picus canus*	LC	L	0	8.11	2.55	S

	Common name	Scientific name	IUCN Red List status	Status of conservation concern	Summer (April–June) (80 checklists)	Monsoon (July–September) (36 checklists)	Winter (October–March) (235 checklists)	Migratory status
43	Common Kestrel	*Falco tinnunculus*	LC	M	6.25	5.41	5.53	YR
44	Eurasian Hobby	*Falco subbuteo*	LC	L	0	0	0.43	S
45	Peregrine Falcon	*Falco peregrinus*	LC	L	0	0	0.85	S
46	Slaty-headed Parakeet	*Psittacula himalayana*	LC	L	43.75	13.51	12.34	YR
47	Long-tailed Minivet	*Pericrocotus ethologus*	LC	M	2.5	2.70	2.13	YR
48	White-browed Shrike-babbler	*Pteruthius aeralatus*	LC	ND	1.25	0	1.28	S
49	White-throated Fantail	*Rhipidura albicollis*	LC	L	0	0	2.13	S
50	Black Drongo	*Dicrurus macrocercus*	LC	L	5	8.12	1.28	YR
51	Ashy Drongo	*Dicrurus leucophaeus*	LC	L	1.25	0	0	S
52	Long-tailed Shrike	*Lanius schach*	LC	M	0	0	1.70	S
53	Eurasian Jay	*Garrulus glandarius*	LC	L	0	0	0.43	S
54	Black-headed Jay	*Garrulus lanceolatus*	LC	L	10	5.41	5.96	YR
55	Yellow-billed Blue Magpie	*Urocissa flavirostris*	LC	L	0	0	0.85	S
56	Red-billed Blue Magpie	*Urocissa erythroryncha*	LC	L	5	8.11	3.83	YR
57	Grey Treepie	*Dendrocitta formosae*	LC	L	13.75	27.03	17.02	YR
58	Spotted Nutcracker	*Nucifraga caryocatactes*	LC	ND	10	29.73	0	S
59	Large-billed Crow	*Corvus macrorhynchos*	LC	L	65	40.54	71.06	YR
60	Yellow-bellied Fantail	*Chelidorhynx hypoxanthus*	LC	L	0	0	0.43	S
61	Grey-headed Canary-flycatcher	*Culicicapa ceylonensis*	LC	M	1.25	2.70	0.43	YR
62	Coal Tit	*Periparus ater*	LC	L	0	0	1.70	S
63	Green-backed Tit	*Parus monticolus*	LC	L	5	10.81	11.91	YR
64	Cinereous Tit	*Parus cinereus*	LC	L	7.5	16.22	10.64	YR
65	Himalayan Black-lored Tit	*Machlolophus xanthogenys*	LC	L	2.5	8.11	4.26	YR
66	Striated Prinia	*Prinia crinigera*	LC	L	68.75	40.54	5.11	YR
67	Dusky Crag Martin	*Ptyonoprogne concolor*	LC	L	2.5	0	0	S
68	Red-rumped Swallow	*Cecropis daurica*	LC	L	10	5.41	1.70	YR
69	Himalayan Bulbul	*Pycnonotus leucogenis*	LC	L	67.5	78.38	61.28	YR
70	Black Bulbul	*Hypsipetes leucocephalus*	LC	L	3.75	16.23	1.70	YR
71	Buff-barred Warbler	*Phylloscopus pulcher*	LC	L	0	0	0.43	S
72	Hume's Warbler	*Phylloscopus humei*	LC	L	0	0	0.85	S
73	Lemon-rumped Warbler	*Phylloscopus chloronotus*	LC	M	2.5	2.70	2.98	YR
74	Common Chiffchaff	*Phylloscopus collybita*	LC	L	0	0	0.43	S
75	Grey-hooded Warbler	*Phylloscopus xanthoschistos*	LC	L	47.5	27.03	29.36	YR
76	Brownish-flanked Bush Warbler	*Horornis fortipes*	LC	L	6.25	5.41	0.851	YR
77	Aberrant Bush Warbler	*Horornis flavolivaceus*	LC	L	0	0	0.43	S
78	Black-throated Tit	*Aegithalos concinnus*	LC	L	13.75	5.41	16.17	YR
79	Whiskered Yuhina	*Yuhina flavicollis*	LC	L	0	0	0.43	S
80	Indian White-eye	*Zosterops palpebrosus*	LC	L	12.5	21.62	3.83	YR
81	Black-chinned Babbler	*Cyanoderma pyrrhops*	LC	L	0	8.11	1.28	S
82	Rusty-cheeked Scimitar Babbler	*Erythrogenys erythrogenys*	LC	L	32.5	29.73	13.62	YR
83	Jungle Babbler	*Argya striata*	LC	L	0	0	0.85	S
84	White-throated Laughingthrush	*Pterorhinus albogularis*	LC	L	0	0	I*	S
85	Rufous-chinned Laughingthrush	*Ianthocincla rufogularis*	LC	L	0	0	0.43	S

	Common name	Scientific name	IUCN Red List status	Status of conservation concern	Summer (April–June) (80 checklists)	Monsoon (July–September) (36 checklists)	Winter (October–March) (235 checklists)	Migratory status
86	Streaked Laughingthrush	*Trochalopteron lineatum*	LC	L	26.25	13.51	20	YR
87	Variegated Laughingthrush	*Trochalopteron variegatum*	LC	L	3.75	0	8.94	S
88	Rufous Sibia	*Heterophasia capistrata*	LC	L	0	2.70	8.09	S
89	Chestnut-tailed Minla	*Actinodura strigula*	LC	L	1.25	0	1.28	S
90	Wallcreeper	*Tichodroma muraria*	LC	L	0	0	0.43	S
91	Chestnut-bellied Nuthatch	*Sitta cinnamoventris*	LC	L	2.5	0	0	S
92	Bar-tailed Treecreeper	*Certhia himalayana*	LC	L	1.25	0	2.98	S
93	Common Myna	*Acridotheres tristis*	LC	L	0	2.70	2.98	S
94	Grey-winged Blackbird	*Turdus boulboul*	LC	L	2.5	2.70	0.43	YR
95	Black-throated Thrush	*Turdus atrogularis*	LC	ND	2.5	0	2.98	S
96	Asian Brown Flycatcher	*Muscicapa dauurica*	LC	M	0	2.7	0	S
97	Verditer Flycatcher	*Eumyias thalassinus*	LC	L	20	5.41	3.83	YR
98	Blue Whistling Thrush	*Myophonus caeruleus*	LC	L	23.75	24.32	33.19	YR
99	Himalayan Bush Robin	*Tarsiger rufilatus*	LC	L	0	0	1.28	S
100	Ultramarine Flycatcher	*Ficedula superciliaris*	LC	L	0	0	0.85	S
101	Plumbeous Water Redstart	*Phoenicurus fuliginosus*	LC	M	I*	0	0	S
102	Blue-capped Redstart	*Phoenicurus coeruleocephala*	LC	L	0	2.70	20.43	S
103	Black Redstart	*Phoenicurus ochruros*	LC	M	0	5.41	0	S
104	Chestnut-bellied Rock Thrush	*Monticola rufiventris*	LC	L	0	2.70	0	S
105	Blue-capped Rock Thrush	*Monticola cinclorhyncha*	LC	L	8.75	5.41	0	S
106	Siberian Stonechat	*Saxicola maurus*	LC	L	20	13.51	1.70	YR
107	Grey Bushchat	*Saxicola ferreus*	LC	L	16.25	5.41	7.66	YR
108	Purple Sunbird	*Cinnyris asiaticus*	LC	L	0	0	0.43	S
109	Scaly-breasted Munia	*Lonchura punctulata*	LC	L	0	0	0.85	S
110	Altai Accentor	*Prunella himalayana*	LC	M	0	0	0.43	S
111	Rufous-breasted Accentor	*Prunella strophiata*	LC	L	1.25	0	1.70	S
112	Black-throated Accentor	*Prunella atrogularis*	LC	M	0	0	2.13	S
113	House Sparrow	*Passer domesticus*	LC	L	16.25	10.81	17.45	YR
114	Russet Sparrow	*Passer cinnamomeus*	LC	L	8.75	0	5.96	S
115	Upland Pipit	*Anthus sylvanus*	LC	M	52.5	32.43	0	S
116	Tree Pipit	*Anthus trivialis*	LC	L	0	0	0.43	S
117	Common Rosefinch	*Carpodacus erythrinus*	LC	L	6.25	0	0	S
118	Pink-browed Rosefinch	*Carpodacus rodochroa*	LC	L	0	0	1.703	S
119	Himalayan white-browed rosefinch	*Carpodacus thura*	LC	M	0	0	0.43	S
120	Plain Mountain Finch	*Leucosticte nemoricola*	LC	L	0	0	3.83	S
121	Yellow-breasted Greenfinch	*Chloris spinoides*	LC	M	0	2.70	2.13	S
122	Fire-fronted Serin	*Serinus pusillus*	LC	L	2.5	0	5.53	S
123	Rock Bunting	*Emberiza cia*	LC	L	1.25	0	19.15	S
124	White-capped Bunting	*Emberiza stewarti*	LC	M	0	0	0.43	S

* Incidental Record(s)
LC—Least Concern | EN—Endangered | NT—Near Threatened | VU—Vulnerable | CR—Critically Endangered.
H—High | M—Moderate | L—Low | ND—Not Determined.
S—Seasonal | YR—Year-round.

information about the species which have been recently split like the Himalayan Owl *Strix nivicolum* (Dixit et al. 2016). It also contains species that have patchy distributions across India (for e.g., Northern Long-eared Owl *Asio otus*) (König & Weick 2010; Grimmett et al. 2011) or western Himalaya (e.g., Cheer Pheasant *Catreus wallichii*, Black Stork *Ciconia nigra*, and Rufous-chinned Laughingthrush *Garrulax rufogularis*). The Red-headed Vulture *Sarcogyps calvus* (BirdLife International 2022) and Koklas Pheasant *Pucrasia macrolopha* (BirdLife International 2016) are two records that additionally include information about the species' upper and lower elevation limits, respectively.

Cheer Pheasant *Catreus wallichii* (Image 1A): Other bird watchers and the author recorded wild individuals 16 times (distinguish from reintroduced individuals based on leg bands) using eBird (eBird 2022b). Sanjeev Kumar (a resident) also photographed three individuals on 30 December 2019. The highest count of birds was 12, recorded on 23 December 2019 (Tiwari 2019e). The absence of this species during monsoon may be either due to local migration of the species from the area or because Cheer Pheasants are less vocally active outside the breeding season (Gaston 1980). This grassland bird is found where areas are disturbed naturally or anthropogenically (Kaul et al. 2022). Cattle grazing and grassland burning in the area help maintain the habitat which supports this species.

Koklass Pheasant *Pucrasia macrolopha*: On 10 December 2019, Thakur (2019) observed a male Koklass Pheasant *Pucrasia macrolopha* about 100 m from the Kanetiya Temple (height c. 2,200 m). According to BirdLife International (2016), this is not far from the species' lowest elevation range. Locals have regularly reported seeing it at an elevation of 300 m higher, suggesting that it may have locally relocated to this area.

Black Stork *Ciconia nigra* (Image 1B) was recorded on 15 May 2020 (Tiwari 2020e). It has isolated records in Himachal Pradesh (Grimmett et al. 2011). After 20 minutes of circling the area, it flew eastward, perhaps on its way back to its breeding grounds.

Cattle Egret *Bubulcus ibis* (Image 1C): Mamta Thakur (resident) recorded one individual in the second week of January 2020. In this study, this species was identified by its yellow beak and differentiated from the Intermediate egret *Ardea intermedia* by its compact body. Though the species has few records from Shimla district (eBird 2022a) and is a resident in altitudinally lower areas of other districts (for e.g., Kangra, Una, Hamirpur, Sirmaur) (Grimmett et al. 2011) this is the only record of the species from the Kanetiya area.

Red-headed Vulture *Sarcogyps calvus* (Image 1F): During the winter, this species was seen flying over the forest located at an altitude of c. 2,000 m on four occasions (01 December 2019 (Tiwari 2019c), 14 December 2019 (Tiwari 2019d), 10 March 2020 (Tiwari 2020a) and 14 February 2020 (Thakur 2020)). This is close to the upper elevation limit of the bird (BirdLife International 2022).

Himalayan Owl *Strix nivicolum*: The species was heard in Seri Village from a *Pistacia integerrima* tree on 4 May 2020 (Tiwari 2020c) and 7 May 2020 (Tiwari 2020d). The distribution of this species is not very well known as it has recently been split from the Tawny Owl *Strix alco* (Dixit et al. 2016).

Northern Long-eared Owls *Asio otus* (Image 2D): Locals and the author recorded 1–4 individuals eight times in the grasslands near Seri village from 4–21 February 2020 (Tiwari & Kumar 2020). The species has erratic records from India (König & Weick 2010; Grimmett et al. 2011) and has only 25 records from the western Himalayan region (Tiwari & Kumar 2020).

Aberrant Bush Warbler *Horornis flavolivaceus*: Sharma (2020) reported the species on 20 February 2020 from the study area. BirdLife International (2017) record its occurrence to the eastern boundary of Himachal Pradesh and Grimmett et al. (2011) do not include Himachal Pradesh in the range of the species. Nevertheless, the species has records from Himachal Pradesh on eBird (eBird 2021a). It has records throughout the Himalayan region, the westernmost from Jammu & Kashmir (year 2019).

Rufous-chinned Laughingthrush *Garrulax rufogularis* (Image 3F): On 30 November 2019 (Tiwari 2019b), four individuals were found in bushes near the foot of a cliff that overhung a piece of grassland at a height of around 1,900 m. On the eBird platform (eBird 2022c), this is the species' fourth report from the Shimla District. The species is widespread in the eastern hills of India and the Himalaya, but its distribution in the western Himalaya is patchy (Grimmett et al. 2011). In Himachal Pradesh, there are more than 100 records, however, they are only found in Kangra (on the state's western border) and the territories around Shimla District (on the state's eastern border).

Wallcreeper *Tichodroma muraria* (Image 3G): One individual was recorded foraging on a rock surface along the road near Seri Village on 23 October 2019 (Tiwari 2019a). This species is found at high altitudes in the Himalaya (c. 3,300–5,000 m) throughout the year but is known to move towards lower elevations (up to c. 600m (eBird 2022e)) during the winter (Kirwan et al. 2020). Therefore, it could have been moving towards lower

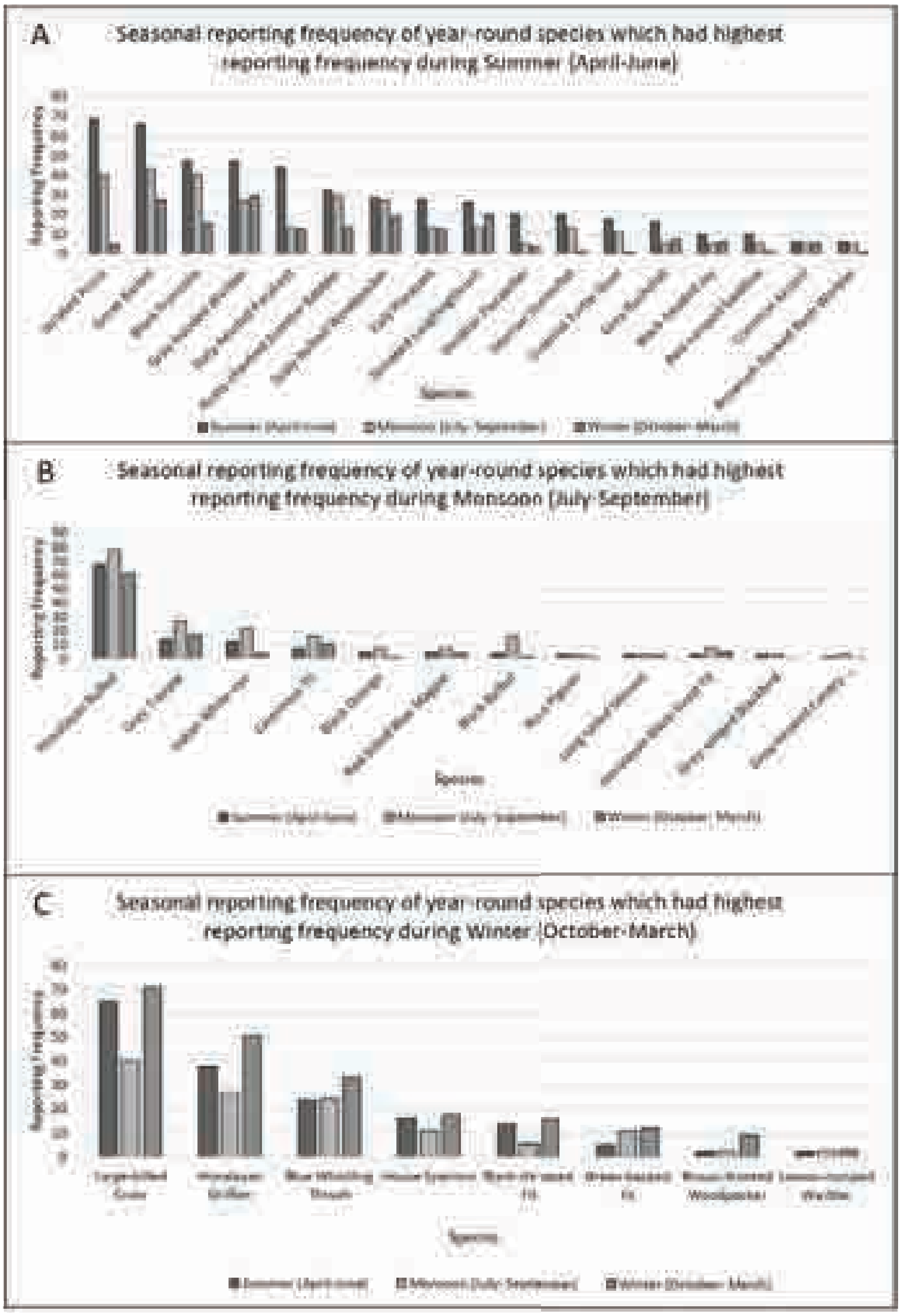

Figure 2. Seasonal reporting frequency of year-round species which had the highest reporting frequency during: A—Summer (April–June) | B—Monsoon (July–September) | C—Winter (October–March).

elevations at the onset of winter in the higher Himalayan region.

Tree Pipit *Anthus trivialis* (Image 3G): Three individuals were recorded in the fields of Seri village (c. 1,850 m) on 20 March 2020 (Tiwari 2020b). This species is a long-distant migrant. It winters (non-breeding season) in peninsular India and migrates to the trans-Himalayas, parts of Europe and North and Central Asia from mid-March to early May (SoIB 2020b; Tyler 2020). Therefore, these individuals could have been moving towards their breeding grounds.

Table 1 presents a comprehensive checklist of bird species recorded in August 2019–2020 along with the IUCN category (IUCN 2020a), category of conservation concern (SoIB 2020a) and reporting frequency across seasons of each species.

DISCUSSION

The environment of the Kanetiya region is shaped by local practices such as grassland burning, cattle grazing, and resource collection. The effects of human activities on bird diversity in the Himalayan terrain can be understood by comparing it to surrounding protected areas. The locals often voluntarily mitigate fires that occur in forest patches. Furthermore, locals protect small swathes of forest known as Devta ka Jungle (sacred groves), which are devoted to regional deities. Customary laws protect these areas from exploitation and destruction Bisht & Ghildiyal 2007; Salick et al. 2007; Anthwal et al. 2010; Singh et al. 2019). These customary laws apply to the forest next to the Kanetiya temple as well, and the land is protected by the locals.

The Cheer Pheasant Reintroduction Programme has been in progress since November 2019 in the designated protected forests and private grasslands close to Seri and Undala villages (IUCN 2020b). The local forest department's conservation program has received backing from the community, which has also taken part. The department could implement additional strategies that involve locals to promote conservation. This may include preparing them to submit simple bird checklists to eBird for monitoring.

Using the citizen science platform eBird, a list of 124 species was created across 3.5 km^2 with 39% of the species recorded from Shimla (eBird 2022d) and 20% of the species from Himachal Pradesh. This variety is brought about by the availability of several habitat types (Somveille et al. 2013; Dixit et al. 2016), elevation fluctuations, and unusual climatic conditions regarding temperature and moisture (Graham et al. 2014). Due to fewer visits to particular environments, some species may have been overlooked because of the non-systematic observations used to create this checklist.

This area is a breeding ground not only for the 37-year-round resident species but also for birds recorded only during the summer. These include Grey Nightjar *Caprimulgus jotaka*, Himalayan Cuckoo *Cuculus saturates*, Wedge-tailed Green Pigeon *Treron sphenurus*, Common Rosefinch *Carpodacus erythrinus*, Lesser Yellownape *Picus chlorolophus*, Dusky Crag Martin *Ptyonoprogne concolor*, Chestnut-bellied Nuthatch *Sitta cinnamoventris*, Booted Eagle *Hieraaetus pennatus* and Ashy Drongo *Dicrurus leucophaeus*. Additionally, birds like Black Stork *Ciconia nigra*, Oriental Honey Buzzard *Pernis ptilorhynchus*, Northern Long-eared Owl *Asio otus* and Tree Pipit *Anthus trivialis* might be using the area as a passage to their breeding grounds as they have incidental records during the summer season.

This area might be serving as a passage to the wintering grounds for some species which were recorded at either a very low reporting frequency or only once at the onset of winter. These include Red-headed Vulture *Sarcogyps calvus*, White-rumped Vulture *Gyps bengalensis*, Aberrant Bush Warbler *Horornis flavolivaceus* and Wallcreeper *Tichodroma muraria*.

Despite recording a high number of birds, some species that are recorded from nearby areas couldn't be recorded during the study period. These include the Green Bee-eater *Merops orientalis*, Blue-tailed Bee-eater *Merops philippinus*, Spot-winged Grosbeak *Mycerobas melanozanthos*, Black-and-yellow Grosbeak *Mycerobas icterioides*, Lesser Cuckoo *Cuculus poliocephalus*, Large Hawk Cuckoo *Hierococcyx sparverioides* and Asian Koel *Eudynamys scolopaceus* (eBird 2022d). While the Purple Sunbird *Cinnyris asiaticus* was recorded during the summer other sunbirds and flowerpeckers couldn't be recorded in any season. I also did not record the Black Kite *Milvus migrans*, which is frequently reported from the Shimla district (eBird 2021b). As per local testimony, the Chukar Partridge *Alectoris chukar* used to occur in the area but became locally extinct 10–15 years ago. Residents had also identified Indian Paradise Flycatcher *Terpsiphone paradise* in previous years, but it was not recorded during the study.

Some species were recorded only near the village houses. These include the Rock Pigeon *Columba livia* which was recorded across all three seasons at very low frequencies (Summer- 2.5, monsoon- 2.70, winter- 0.85) and House Sparrow *Passer domesticus* which was recorded at slightly higher frequencies across seasons (summer- 16.25, monsoon- 10.81, winter- 17.45). The Common Myna *Acridotheres tristis* was also recorded exclusively near village houses in the monsoon (2.70) and winter (2.98).

Most species recorded across all three seasons were rare (recorded with a low reporting frequency) (Figure 3). This pattern is seen in many other studies conducted across various ecosystems (Brown 1984).

Though such non-systematically collected information is valuable (Barnes et al. 2015), the scope of studies based on opportunistic observations can be limited (Snäll et al. 2011; Bird et al. 2014; Henckel et al. 2020). Reporting frequency is a function of abundance and detectability of a species (SoIB 2020a), but as detectability of a species varies among observers with different abilities for different species, it cannot be used to assess the change in population sizes of birds. Therefore, this study only provides a baseline index of abundance across seasons.

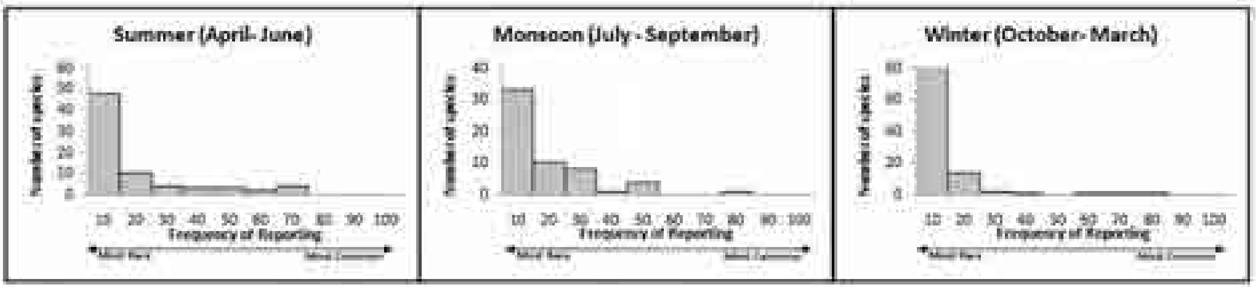

Figure 3. Number of rare (recorded with a low reporting frequency) and common (recorded with a high reporting frequency) bird species recorded across seasons.

A more systematic study based on consistent sampling protocol and effort can provide better information on the change in population of different species and can also be used to confirm true absences accurately (Thompson 2002).

A bird monitoring scheme focusing on unprotected areas can be developed by training bird watchers across the Himalaya to consistently record birds. This will require a simple and yet strict sampling design. Bird Count India (2021) is executing a similar effort at the national level as the Patch Monitoring Project. Such systemic surveys based on community participation can be more widespread and less resource-intensive (Neate-Clegg et al. 2020). They will also help create awareness and aid in conservation.

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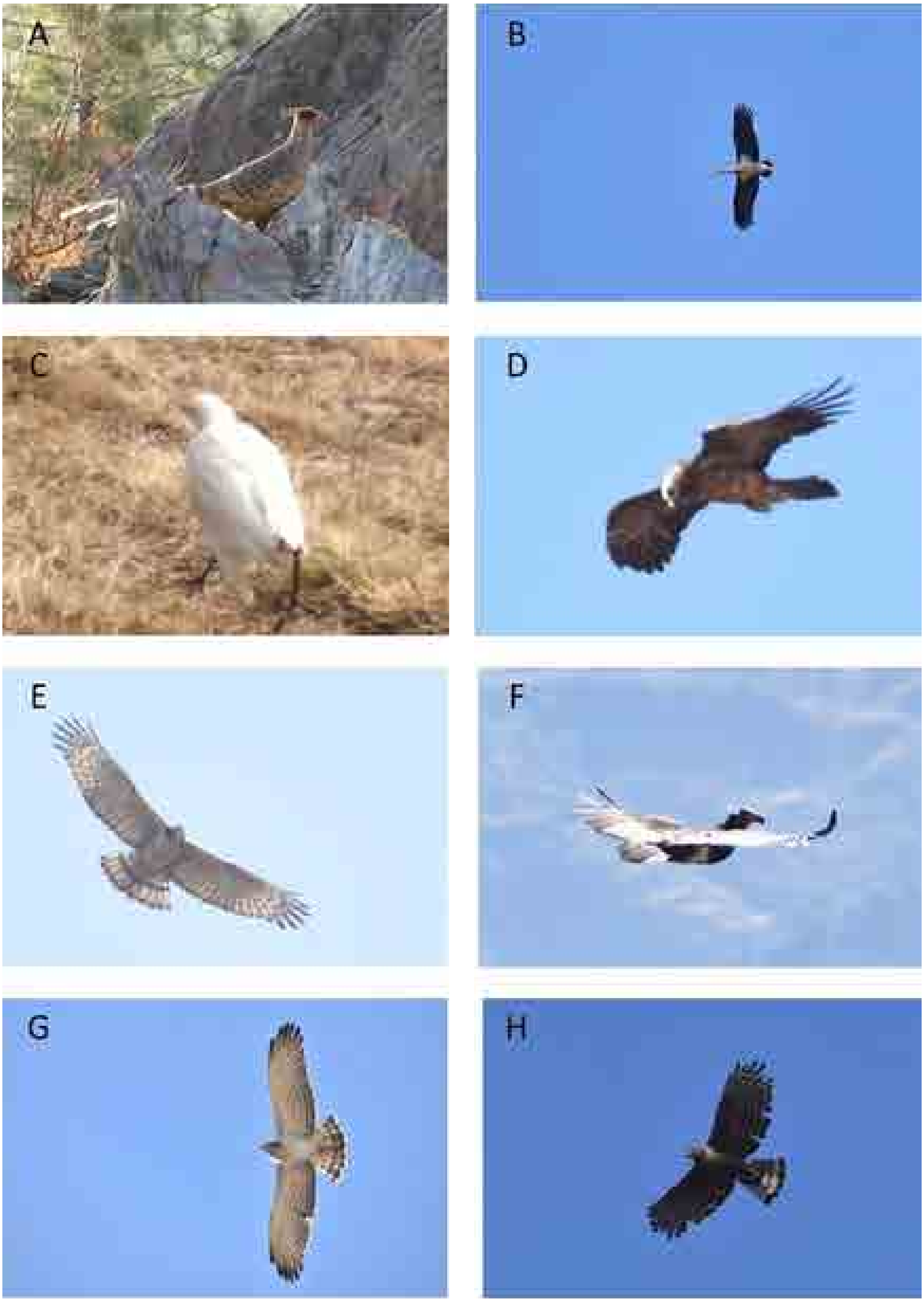

Image 1. Photographic records of some species from the Kanetiya Area: A—Cheer Pheasant *Catreus wallichii* | B—Black Stork *Ciconia nigra* | C—Cattle egret *Bubulcus ibis* | D—Bearded Vulture *Gypaetus barbatus* | E—Oriental Honey Buzzard *Pernis ptilorhynchus* | F—Red-headed Vulture *Gypaetus barbatus* | G—Short-toed Snake Eagle *Circaetus gallicus* | H—Mountain Hawk Eagle *Nisaetus nipalensis*. © A—Sanjeev Kumar | C—Mamta Thakur | Others—Samakshi Tiwari.

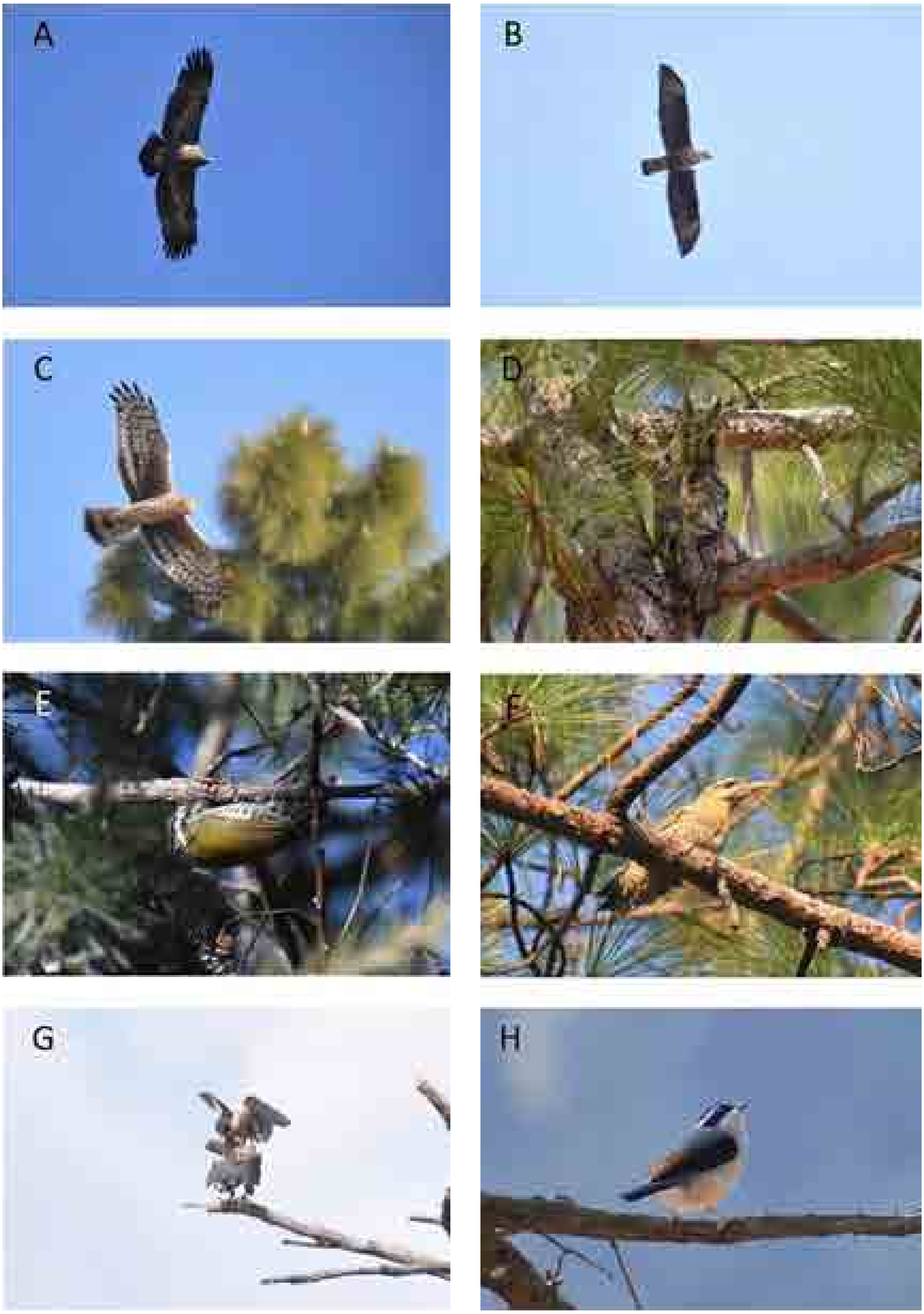

Image 2. Photographic records of some species from the Kanetiya Area: A—Steppe Eagle *Aquila nipalensis* | B—Bonelli's Eagle *Aquila fasciata* | C—Hen Harrier *Circus cyaneus* | D—Northern Long-eared Owl *Asio otus* | E—Speckled Piculet *Picumnus innominatus* | F—Scaly-bellied Woodpecker *Picus squamatus* | G—Eurasian Hobby *Falco subbuteo* | H—White-browed Shrike-babbler *Pteruthius aeralatus*. © Samakshi Tiwari.

Image 3. Photographic records of some species from the Kanetiya Area: A—Long-tailed Shrike *Lanius schach* | B—Red-billed Blue Magpie *Urocissa erythroryncha* | C—Spotted Nutcracker *Nucifraga caryocatactes* | D—Coal Tit *Periparus ater* | E—Lemon-rumped Warbler *Phylloscopus chloronotus* | F—Rufous-chinned Laughingthrush *Ianthocincla rufogularis* | G—Wallcreeper *Tichodroma muraria* | H—Chestnut-bellied Nuthatch *Sitta cinnamoventris*. © Samakshi Tiwari.

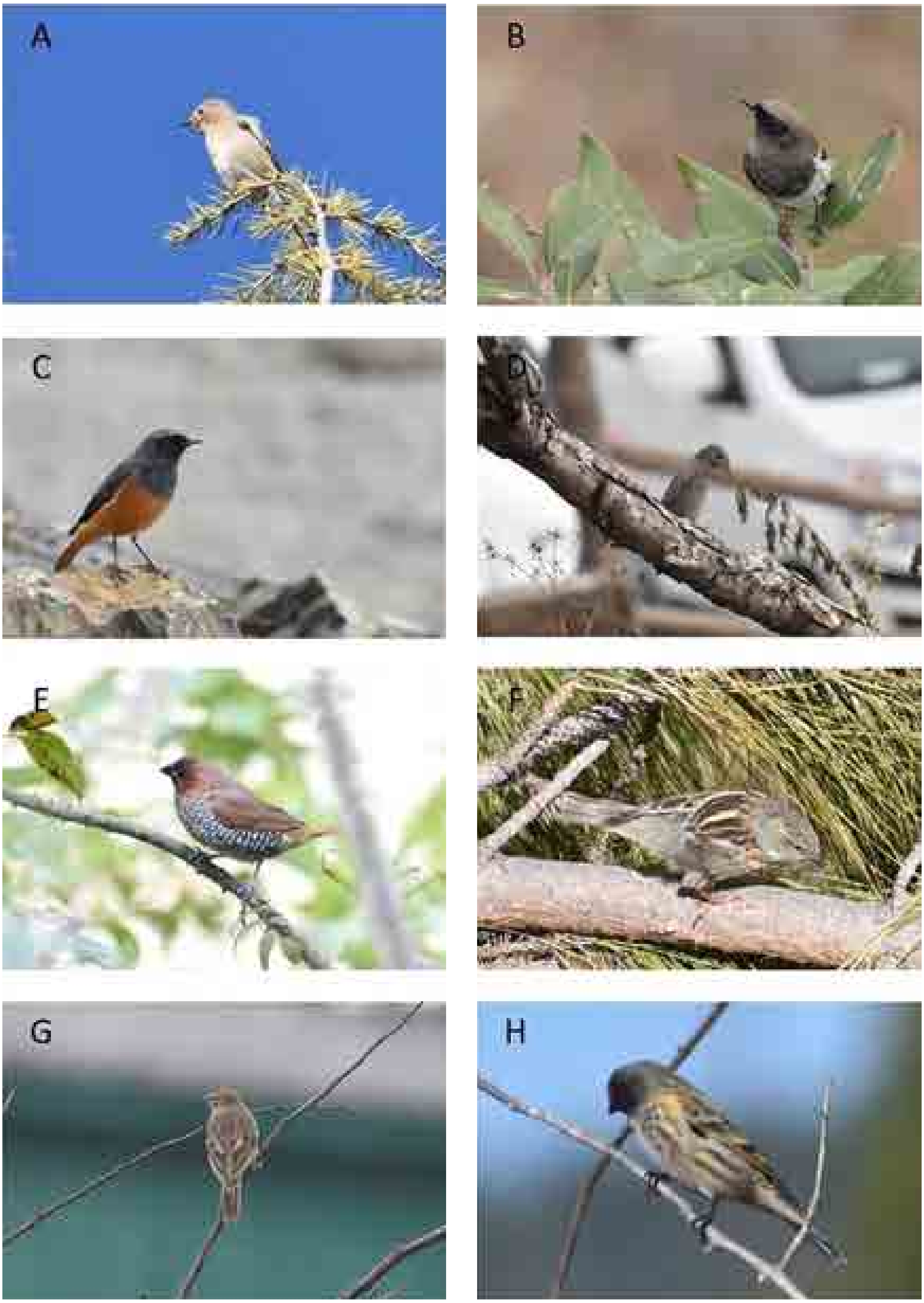

Image 4. Photographic records of some species from the Kanetiya Area: A—Asian Brown Flycatcher *Muscicapa dauurica* | B—Blue-capped Redstart *Phoenicurus coeruleocephala* | C—Black Redstart *Phoenicurus ochruros* | D—Chestnut-bellied Rock Thrush *Monticola rufiventris* | E—Scaly-breasted Munia *Lonchura punctulata*| F—Altai Accentor *Prunella himalayana* | G—Tree Pipit *Anthus trivialis* | H—Fire-fronted Serin *Serinus pusillus*. © Samakshi Tiwari.

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Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23545–23556

ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)

https://doi.org/10.11609/jott.8203.15.7.23545-23556

#8203 | Received 21 November 2022 | Final received 13 June 2023 | Finally accepted 22 June 2023

COMMUNICATION

A preliminary assessment of Odonata (dragonflies & damselflies) across an elevation gradient – insights from Shiwaliks to Alpines, northwestern Himalaya, India

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Abstract: Understanding the species distribution and richness along an environmental gradient helps identify hotspots and prioritize conservation efforts at landscape scale. This is more effective for the species that are indicators of environmental change, such as odonates. As the information about the distribution of this group of insects is scarce in Jammu & Kashmir, their documentation assumes a greater significance. Here, we present a checklist of odonate species from 23 sites across diverse landscapes in subtropical, temperate, and alpine ecosystems over an elevational gradient of 3,700 m in Jammu division. We recorded 63 species from 39 genera and 11 families, four Anisoptera and seven Zygoptera. The most represented families were Libellulidae (15 genera & 29 species) and Coenagrionidae (five genera & 10 species). The preliminary surveys resulted in addition of 24 new species to the Odanata fauna of Jammu & Kashmir, including three new to the northwestern Himalaya. The study underlines that even opportunistic records are useful in understanding the distribution range and delineating the potential habitats of odonates. The study calls for intensive odonate surveys to better understand their distribution and ecology in hitherto less explored region in the northwestern Himalaya.

Keywords: Amphibiotic insects, Anisoptera, Greater Himalaya, mountains, Pir-Panjal, puddles, stream off shoots, summer ditches, Zygoptera

अमूर्त: पर्यावरण ढाल दे कन्नै-कन्नै प्रजातियें दे बंड ते समृद्धि गी समझने कन्नै हॉटस्पॉट दी पन्छान ते लैंडस्केप पैमाने पर संरक्षण दे प्रयासें गी प्राथमिकता देने च मदद मिलदी ऐ। एह् उनें प्रजातियें आस्तै मता असरदार ऐ जेह् ड़ियां पर्यावरण च बदलाव दे संकेतक न, जि'यां ओडोनेट। चूंकि जम्मू-कश्मीर च कीड़े-मकोड़े दे इस समूह दे बंड बारै जानकारी घट्ट ऐ, इस करी उंदे दस्तावेजें दा मता महत्व ऐ। इत्थें, अस उप-उष्णकटिबंधीय, समशीतोष्ण ते अल्पाइन पारिस्थितिकी प्रणाली च बक्ख-बक्ख परिदृश्यें च 23 थाहें थमां ओडोनाटा प्रजातियें दी जांच सूची पेश करदे आं। असें 39 जीनस ते 11 परिवारें थमां 63 प्रजातियां, चार एनिसोप्टेरन ते 7 ज़ाइगोप्टेरन दर्ज कीतियां ! सारें शा मते प्रतिनिधित्व आह्‌ले परिवार लिबेलुलिडी हे जिंदे च 15 जीनस ते 29 प्रजातियां ते कोएनग्रिओनिडी (5 जीनस ते 10 प्रजातियां) शामल हे ! सर्वेक्षणें दे नतीजे च जम्मू-कश्मीर दे ओडानाटा जीव-जंतुएं च 24 नमीं प्रजातियां बी शामल कीतियां गेइयां जिंदे च उत्तर-पश्चिमी हिमालय च त्रै नमीं प्रजातियां बी शामल न। अध्ययन इस गल्लै गी रेखांकित करदा ऐ जे अवसरवादी रिकार्ड बी वितरण सीमा गी समझने ते ओडोनेट दे संभावित आवासें गी रेखांकित करने च उपयोगी न। इस अध्ययन च उत्तर-पश्चिमी हिमालय च अजें तगर कम खोजे गेदे इलाके च उंदे बंड ते पारिस्थितिकी गी बेहतर तरीके कन्नै समझने लेई गहन ओडोनेट सर्वेक्षण दा आह्‌वान कीता गेआ ऐ।

Editor: Pankaj Koparde, MIT World Peace University, Pune, India. **Date of publication:** 26 July 2023 (online & print)

Citation: Sharma, N., D. Singh, S. Sharma & A. Ansari (2023). A preliminary assessment of Odonata (dragonflies & damselflies) across an elevation gradient – insights from Shiwaliks to Alpines, northwestern Himalaya, India. *Journal of Threatened Taxa* 15(7): 23545–23556. https://doi.org/10.11609/jott.8203.15.7.23545-23556

Funding: The surveys were partly funded by Department of Wildlife Protection, Govt. of Jammu and Kashmir, and Ministry of Environment, Forest and Climate Change, Govt. of India.

Competing interests: The authors declare no competing interests.

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Author contributions: NS, SS and DS conducted the field surveys and drafted the manuscript; NS and SS conceptualized, designed, and executed the surveys and wrote the manuscript; AA analysed the data, while SS communicated with the journal.

Acknowledgements: The authors would like to express their gratitude to Rector, Bhaderwah Campus, University of Jammu, for his administrative support during the surveys. Department of Wildlife Protection, Government of the UT of Jammu & Kashmir and Ministry of Environment, Forest and Climate Change, Govt. of India are thanked for partly funding the surveys. The authors gratefully appreciate the help rendered by Asha, Anil, Aasma and Vandana for assisting with field surveys and Muzaffar A Kichloo for the preparation of map.

INTRODUCTION

The Himalayan Odonata fauna comprises of 257 species in 112 genera and 18 families with 34 species endemic to the Himalayas (Subramaniam & Babu 2018). Though well-documented in the neighbouring states of Himachal Pradesh and Uttarakhand, the information on the distribution of Odonata in Jammu & Kashmir is scanty. The earlier accounts of Odonata from Jammu & Kashmir dates back to the records of Abott (Calvert 1899), Fraser (1933, 1934, 1936) followed by a few checklists (Singh & Baijal 1954; Asahina 1978; Kumar & Prasad 1981; Carfi et al. 1983; Kumar 1983; Lahiri & Das 1991; Dar et al. 2002; Mitra 2003). Recently, a few surveys have been conducted to describe the diversity and distribution of odonates of Jammu & Kashmir (Subramanian & Babu 2018; Sheikh et al. 2020; Riyaz & Sivasankaran 2021; Quereshi et al. 2022; Kumar et al. 2022). Singh (2022) described 65 species from Jammu & Kashmir based on available literature and the online curated website Odonata of India (https://www.indianodonata.org/) published until 2022.

Geographically, the union territory of Jammu & Kashmir comprises two regions, Jammu & Kashmir characterized by five distinct physiographic units. The Jammu region of Jammu & Kashmir offers a wide range of habitats from the alluvial plains of the Ravi and Chenab rivers in the south to the moderately elevated Shiwaliks, Pir-Panjal, and Greater Himalaya northwards, bordering Kashmir in the north and Ladakh in the north-east. Documenting odonate fauna from such regions becomes important as it helps fill the knowledge gap about distribution of species, which may have conservation implications. To gain a better understanding of the spatial distribution of odonates in the region, we conducted preliminary surveys in seasonal and perennial water bodies in parts of alluvial plains, sub-tropics, lesser, and the Greater Himalaya spanning a vast elevational gradient ranging from 260–3,960 m. The baseline information obtained on the abundance and distribution of 63 species of odonates for the region will be useful for monitoring the health of aquatic ecosystems on spatial and temporal scales.

METHODS

We sampled the adult dragonflies in 23 stations: eleven in the subtropics, nine in temperate, and three in alpine habitats in Rajouri, Jammu, Udhampur, Kathua,

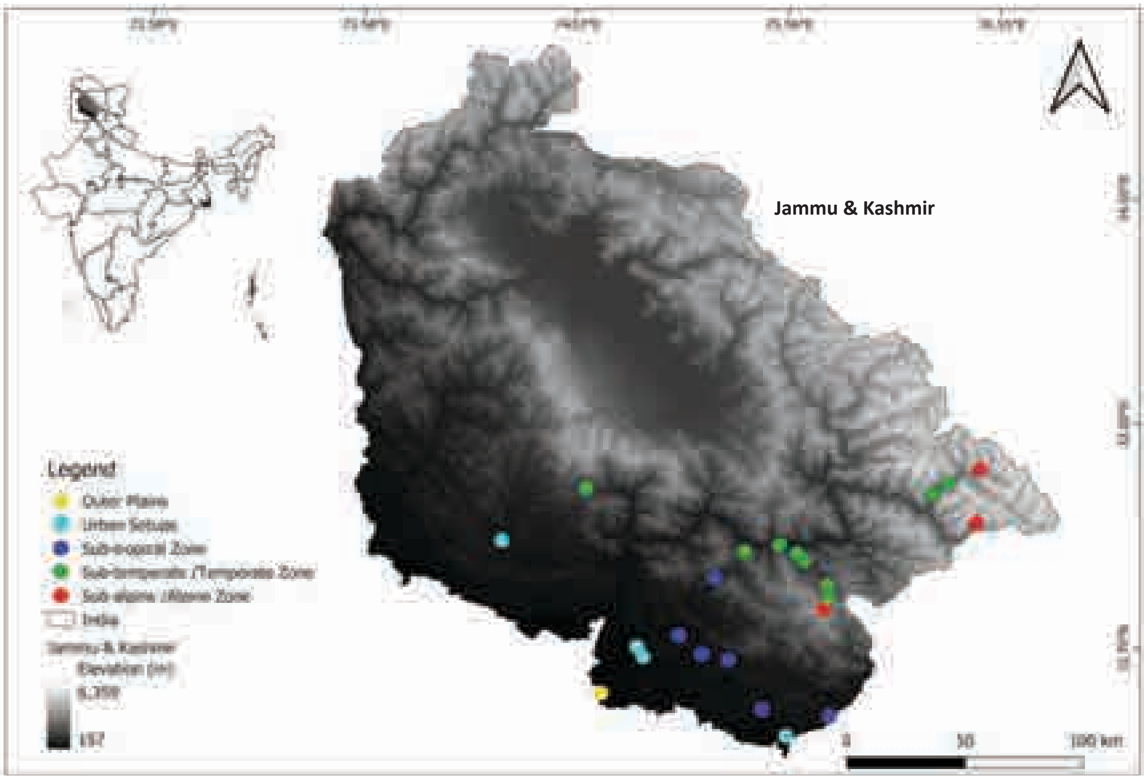

Figure 1. The sampling sites in three climatic zones (subtropical, subtemperate / temperate and alpine) in the study area. The outer plains and urban setups lie in the subtropical zone, a part of Jammu Shiwaliks.

Doda, and Kishtwar districts of Jammu division in the union territory of Jammu & Kashmir. The subtropical habitats included the alluvial plains and hills and ridges of Jammu Shiwaliks (250–1,150 m). Characterized by dry climate and soils with low water retention capacity, most of the sampled habitats included seasonal ponds, roadside ditches, and streams. The subtemperate and temperate ecosystems in the lesser Himalayas were scanned for forested perennial channels, rivulets, and streams in an elevation range of 1,150 to 3,000 m, while sub alpines and alpines included springs, minor streams, summer ditches, and swamps (3,200–4,200 m) in parts of the Greater Himalaya bordering the Zanskar region of Ladakh (Table 1, Image 1). During a two-year sampling period, we followed an opportunistic sampling strategy and visited all stations twice during summer to monsoon (end of May to mid-September) except for the alpines which were too far apart. The individuals were not

Table 1. Spatial attributes of sampling locations, including geomorphological features and the degree of disturbance.

Climate zones	Sampling sites / District	Geo co-ordinates *	Elevation (in m) *	Habitat description	Disturbance
Subtropical zone	1.Gharana and associated wetlands in agricultural landscape, Jammu	32.540°N, 74.690° E	260	A vast agriculture landscape comprised of a protected wetland, marshlands, ditches, channels, and paddy fields	High
	2.GGM Science College, Jammu	32.724° N, 74.851° E	302	Botanical Garden and college lawns	Moderate
	3. Kathua town, Kathua	32.367 ° N, 75.525 ° E	318	Urban drain dissecting the twon	High
	4.Trikutanagar, Jammu	32.685° N, 74.879° E	320	Urban storm water drain	High
	5. Nowshera, Rajouri	33.149 ° N, 74.234 ° E	543	Urban drain at the confluence with a perennial stream	High
	6. Jasrota WLS / Ujh Barrage, Kathua	32.474° N, 75.417° E	382	A lacustrine ecosystem surrounded with plantations, farmlands and habitations.	Low
	7. Thein Conservation Reserve, Kathua	32.446° N, 75.721° E	518	A terrestrial protected area bounded by a reservoir eastward.	Low
	8. Surinsar, Jammu	32.770° N, 75.041° E	605	Medium sized lake, seasonal ponds and channels	Moderate
	9. Battal, Udhampur	32.672° N, 75.264° E	630	Streams, roadside water channels and ditches	Low
	10. Mansar, Udhampur	32.696° N, 75.145° E	662	Large water body, ponds, and ditches	Moderate
	11. Samroli, Udhampur	33.002° N, 75.206° E	845	Seasonal and perennial streams, ditches, and roadside drains	Moderate
Subtemperate / Temperate zone	12. Pranoo, Doda	33.097° N, 75.580° E	1210	Neeru stream, main channel	Low
	13. Bhalla, Doda	33.068° N, 75.613° E	1270	Neeru stream and tributaries	Low
	14. Khellani, Doda	33.132° N, 75.500° E	1350	Streams, roadside water channels, and springs	Low
	15. Batote, Doda	33.106° N, 75.341° E	1430	Roadside springs, water channels and rivulets	Moderate
	16. Phalni, Rajouri	33.361°N, 74.621° E	1440	A fish farm housing Indian Major Carps adjacent to trout raceways of state fisheries department.	Low
	17. Bhaderwah, Doda	32.969° N, 75.718° E	1714	Springs, water channels and ditches in Bhaderwah Campus, seasonal channels	Low
	18. Kundail, Kishtwar	33.331° N, 76.204° E	2075	Bhot stream and springs	Low
	19. Thanthera, Doda	32.918° N, 75.723° E	2155	Basti stream and roadside springs	Low
	20. Chasoti, Kishtwar	33.374° N, 76.275° E	2356	A typical mountain village with terrace farmlands	Low
Subalpine / Alpine zone	21. Suncham, Kishtwar	32.429° N, 76.410° E	3260	Bhot stream, its tributary Hagshu, springs and channels.	Low
	22. Tun, Kishtwar	33.208° N, 76.396° E	3345	Sansari stream, seasonal ditches, and springs	Low
	23. Kailash Lake, Doda	32.871° N, 75.699° E	3960	Natural springs and rivulets	Low

* Geo co-ordinates and elevation taken as the centre point of each 1.5–2 km^2 grids sampled.

counted for abundance and density estimates, however, the frequency of sightings was taken into account for computing the relative frequency. The odonates were categorized as very common (sighted during 75–100 % of the sampling), common (sighted between 50–75 % times), occasional (observed between 25–50 %), and rare (sighted below 25% times) following Adarsh et al. (2014). All the field visits were conducted between 1000 h to 1200 h, when the adult odonates are most active. The individual odonates were photographed and identified to the species level referring to the field guides (Subramanian 2005, 2009; Kiran & Raju 2013; Singh 2022) and curated online platforms like Odonata of India website (https://www.indianodonata.org/). No specimens were, however, collected during the surveys. The species have been enlisted following the systematic arrangement and taxonomy of Subramanian et al. (2018) and Kalkman et al (2020).

RESULTS

A total of 63 odonates (40 dragonflies and 23 damselflies) were recorded from the study area. These belonged to 39 genera and 11 families, four anisopterans and seven zygopterans (Table 2, Figure 2, Images 1–63). In terms of habitat sharing, 50 species were exclusively found to be associated with one of the three ecosystems studied, indicating their limited geographical distribution. The sub-tropical ecosystems harboured high richness (SR = 46) accounting for 73% of the total, followed by temperate (SR = 28, 43%) and alpine (SR = 5, 8%). Fourteen species were found to be common across subtropical and temperate ecosystems, whereas temperate and alpine shared only two species, *Cordulegaster brevistigma* and *Orthetrum internum*. Families Calopterygidae, Chlorocyphidae, Chlorocyphidae, and Platycnemididae were confined to subtropical habitats, while Gomphidae, Libellulidae, Coenagrionidae, Euphaeidae, Lestidae, and Synlestidae exhibited affinities for both sub-tropical and temperate climate (Figure 3).

The occurrence data (relative frequency) during the study period shows that 48 species (76%) belonged to occasional and rare (n = 24, each) category. Eleven species were found common and four very common. *Orthetrum pruinosum*, *O. triangulare*, and *Sympetrum commixtum* among the Anisoptera and *Amphiallagma parvum* among the Zygoptera were the most commonly encountered species during the current sampling. In all, 60 species are classified as 'Least Concern' by the IUCN, while three species have not yet been evaluated for their threat status (Table 2). Families Aeshnidae and Libellulidae are found in all three climatic zones, occupying a greater elevational range than other Anisoptera families (Figure 3). Family Cordulegastridae comprising a solitary taxon *Cordulegaster brevistigma* was restricted to temperate and alpine zones, whilst members of the family Gomphidae were restricted to subtropical and temperate regions. Most of

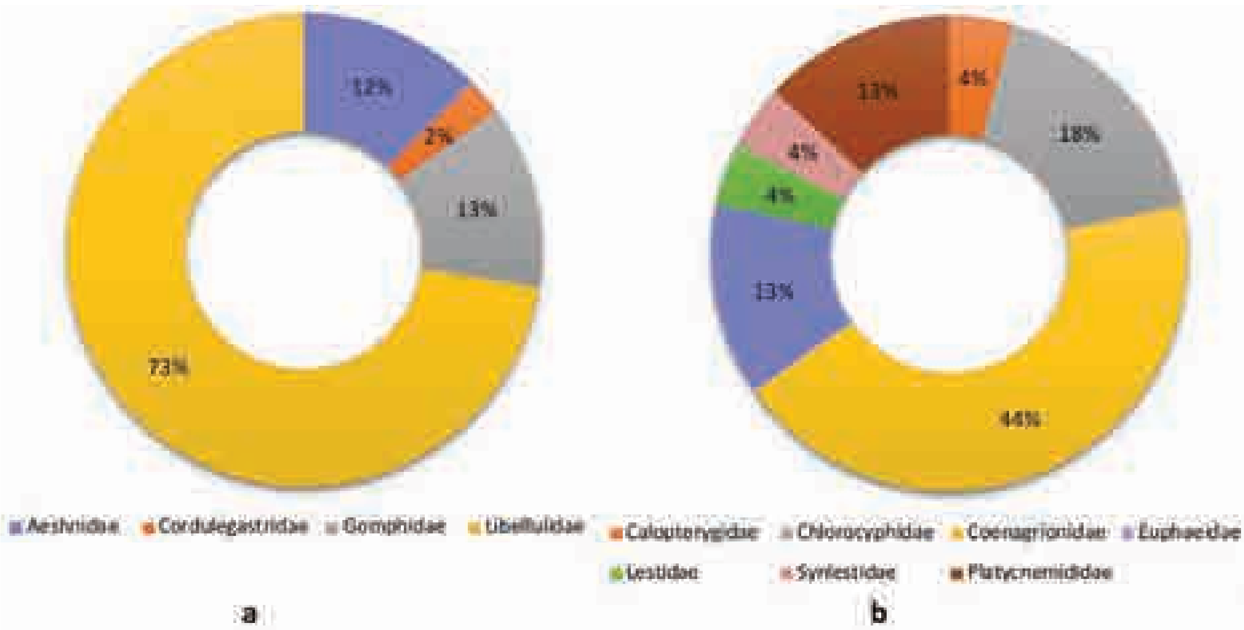

Figure 2. The species observed in different families: a—Anisoptera | b—Zygoptera.

Table 2. Checklist of odonates in the three distinct ecosystems in Jammu division of Jammu & Kashmir depicting distribution, relative frequency and threat status.

	Suborder / Family / Species	Distribution			Relative frequency	IUCN Red List status	Image number
		ST	TM	AL			
Suborder : Anisoptera (Dragonflies)							
Family : Aeshnidae							
1.	*Aeshna juncea* (Bartenef, 1929)	-	-	+	RA	LC	1
2.	*Anax immaculifrons* (Rambur, 1842)	+	-	-	OC	LC	2
3.	*Anax indicus* (Lieftinck, 1942)*	+	-	-	RA	LC	3
4.	*Anax nigrolineatus* (Fraser, 1935)*	-	+	-	OC	LC	4
5.	*Anax parthenope* (Selys, 1839)	-	+	-	RA	LC	5
Family : Cordulegastridae							
6.	*Cordulegaster brevistigma* (Selys, 1854)	-	+	+	OC	LC	6
Family : Gomphidae							
7.	*Anisogomphus bivittatus* (Selys, 1854)*	-	+	-	OC	LC	7
8.	*Davidius davidii* (Selys, 1878)**	-	+	-	RA	LC	8
9.	*Ictinogomphus rapax* (Rambur, 1842)*	+	-	-	OC	LC	9
10.	*Ophiogomphus reductus* (Calvert, 1898)	-	+	-	OC	LC	10
11.	*Paragomphus lineatus* (Selys, 1850)	+	+	-	OC	LC	11
Family : Libellulidae							
12	*Acisoma panorpoides* (Rambur, 1842)	+	-	-	RA	LC	12
13	*Brachythemis contaminata* (Fabricius, 1793)	+	-	-	OC	LC	13
14	*Brachydiplax sobrina* (Rambur, 1842)*	+	-	-	RA	LC	14
15	*Bradinopyga geminata* (Rambur, 1842)	+	-	-	OC	LC	15
16	*Crocothemis erythraea* (Brullé, 1832)	+	-	-	OC	LC	16
17	*Crocothemis servilia* (Drury, 1770)	+	+	-	CO	LC	17
18	*Diplacodes lefebvrii* (Rambur, 1842)	+	-	-	OC	LC	18
19	*Diplacodes nebulosa* (Fabricius, 1793)*	+	-	-	RA	LC	19
20	*Libellula quadrimaculata* (Linnaeus, 1758)	-	+	-	RA	LC	20
21	*Neurothemis tullia* (Drury, 1773)	+	-	-	RA	LC	21
22	*Orthetrum cancellatum* (Linnaeus, 1758)	-	+	-	RA	LC	22
23	*Orthetrum glaucum* (Brauer, 1865)	-	+	-	OC	LC	23
24	*Orthetrum internum* (McLachlan, 1894)	-	+	+	CO	NE	24
25	*Orthetrum luzonicum* (Brauer, 1868)	-	+	-	CO	LC	25
26	*Orthetrum pruinosum* (Burmeister, 1839)	+	+	-	VC	LC	26
27	*Orthetrum sabina* (Drury, 1770)	+	-	-	CO	LC	27
28	*Orthetrum taeniolatum* (Schneider, 1845)*	+	-	-	RA	LC	28
29	*Orthetrum triangulare* (Selys, 1878)	+	+	-	VC	LC	29
30	*Palpopleura sexmaculata* (Fabricius, 1787)	+	+	-	CO	LC	30
31	*Pantala flavescens* (Fabricius, 1798)	+	-	-	OC	LC	31
32	*Rhyothemis triangularis* (Kirby, 1889)	+	-	-	RA	LC	32
33	*Rhyothemis variegata* (Linnaeus, 1763)	+	-	-	OC	LC	33
34	*Sympetrum commixtum* (Selys, 1884)	-	-	+	VC	LC	34
35	*Sympetrum fonscolombii* (Selys, 1840)	-	+	-	RA	LC	35
36	*Sympetrum speciosum* (Oguma, 1915)*	-	-	+	CO	NE	36
37	*Tramea transmarina* (Selys, 1878)**	-	+	-	OC	LC	37

	Suborder / Family / Species	Distribution			Relative frequency	IUCN Red List status	Image number
		ST	TM	AL			
38	*Tramea virginia* (Rambur, 1842)	+	+	-	OC	LC	38
39	*Trithemis aurora* (Burmeister, 1839)	+	-	-	OC	LC	39
40	*Trithemis festiva* (Rambur, 1842)	+	+	-	CO	LC	40
Suborder : Zygoptera (Damselflies)							
Family : Calopterygidae							
41	*Neurobasis chinensis* (Linnaeus, 1758)	+	-	-	RA	LC	41
Family : Chlorocyphidae							
42	*Aristocypha trifasciata* (Selys, 1853)*	+	-	-	OC	LC	42
43	*Aristocypha quadrimaculata* (Selys, 1853)	+	-	-	CO	LC	43
44	*Paracypha unimaculata* (Selys, 1853)	+	-	-	RA	LC	44
45	*Libellago lineata* (Burmeister, 1839)	+	-	-	OC	LC	45
Family : Coenagrionidae							
46	*Amphiallagma parvum* (Selys, 1876)*	+	+	-	VC	LC	46
47	*Agriocnemis pygmaea* (Rambur, 1842)*	+	-	-	RA	LC	47
48	*Ceriagrion coromandelianum* (Fabricius, 1798)	+	-	-	CO	LC	48
49	*Ischnura forcipata* (Morton, 1907)	+	+	-	CO	LC	49
50	*Ischnura rubilio* (Selys, 1876)	+	+	-	OC	NE	50
51	*Pseudagrion decorum* (Rambur, 1842)*	+	-	-	RA	LC	51
52	*Pseudagrion hypermelas* (Selys, 1876)*	+	-	-	RA	LC	52
53	*Pseudagrion microcephalum* (Rambur, 1842)*	+	-	-	RA	LC	53
54	*Pseudagrion rubriceps* (Selys, 1876)	+	+	-	OC	LC	54
55	*Pseudagrion spencei* (Fraser, 1922)*	+	-	-	OC	LC	55
Family : Euphaeidae							
56	*Anisopleura comes* (Hagen,1880)*	+	+	-	RA	LC	56
57	*Anisopleura lestoides* (Selys,1853)*	-	+	-	OC	LC	57
58	*Bayadera indica* (Selys, 1853)*	+	-	-	RA	LC	58
Family : Lestidae							
59	*Lestes dorothea* (Fraser, 1924)*	+	+	-	OC	LC	59
Family : Synlestidae							
60	*Megalestes major* (Selys, 1862)*	+	+	-	CO	LC	60
Family : Platycnemididae							
61	*Calicnemia imitans* (Lieftinck, 1948)**	+	-	-	RA	LC	61
62	*Copera marginipes* (Rambur, 1842)	+	-	-	RA	LC	62
63	*Drepanosticta carmichaeli* (Laidlaw, 1915)*	+	-	-	RA	LC	63

+—presence | -—absence | ST—Sub-tropical | TM—Subtemperate / Temperate zone | AL—Alpine zone | OC—Occasional | CO—Common | VC—Very Common | RA—Rare | LC—Least Concern | NE—Not Evaluated | *—Addition to the Odonata fauna of Jammu & Kashmir | **—Addition to the Odonata fauna of northwestern Himalaya, India.

the Zygoptera families occupied subtropical regions, although a few extended their range into sub-temperate and temperate zones (Figure 3).

Twenty-four (10 Anisoptera and 14 Zygoptera) among the 63 species encountered during the surveys are reported for the first time in Jammu & Kashmir, including three new to the northwestern Himalaya. These include *Anax indicus*, *A. nigrolineatus*, *Anisogomphus bivittatus*, *Ictinogomphus rapax*, *Brachydiplax sobrina*, *Diplacodes nebulosa*, *Orthetrum taeniolatum*, *Sympetrum speciosum*, *Aristocypha trifasciata*, *Amphiallagma parvum*, *Agriocnemis pygmaea*, *Pseudagrion decorum*, *P. hypermelas*, *P. microcephalum*, *P. spencei*, *Anisopleura comes*, *A. lestoides*, *Bayadera indica*,

Images 1–18. 1—*Aeshna juncea* | 2—*Anax immaculifrons* | 3—*Anax indicus* | 4—*Anax nigrolineatus* | 5—*Anax parthenope* | 6—*Cordulegaster brevistigma* | 7—*Anisogomphus bivittatus* | 8—*Davidius davidii* | 9—*Ictinogomphus rapax* | 10—*Ophiogomphus reductus* | 11—*Paragomphus lineatus* | 12—*Acisoma panorpoides* | 13—*Brachythemis contaminata* | 14—*Brachydiplax sobrina* | 15—*Bradinopyga geminata* | 16—*Crocothemis erythraea* | 17—*Crocothemis servilia* | 18—*Diplacodes lefebvrii*.

Images 19–36. 19—***Diplacodes nebulosa*** | 20—***Libellula quadrimaculata*** | 21—***Neurothemis tullia*** | 22—***Orthetrum cancellatum*** | 23—***Orthetrum glaucum*** | 24—***Orthetrum internum*** | 25—***Orthetrum luzonicum*** | 26—***Orthetrum pruinosum*** | 27—***Orthetrum sabina*** | 28—***Orthetrum taeniolatum*** | 29—***Orthetrum triangulare*** | 30—***Palpopleura sexmaculata*** | 31—***Pantala flavescens*** | 32—***Rhyothemis triangularis*** | 33—***Rhyothemis variegata*** | 34—***Sympetrum commixtum*** | 35—***Sympetrum fonscolombii*** | 36—***Sympetrum speciosum.***

Images 37—54. 37—*Tramea transmarina* | 38—*Tramea virginia* | 39—*Trithemis aurora* | 40—*Trithemis festiva* | 41—*Neurobasis chinensis* | 42—*Aristocypha trifasciata* | 43—*Aristocypha quadrimaculata* | 44—*Paracypha unimaculata* | 45—*Libellago lineata* | 46—*Amphiallagma parvum* | 47—*Agriocnemis pygmaea* | 48—*Ceriagrion coromandelianum* | 49—*Ischnura forcipata* | 50—*Ischnura rubilio* | 51—*Pseudagrion decorum* | 52—*Pseudagrion hypermelas* | 53—*Pseudagrion microcephalum* | 54—*Pseudagrion rubriceps.*

Images 55–63. 55—*Pseudagrion spencei* | 56—*Anisopleura comes* | 57—*Anisopleura lestoides* | 58—*Bayadera indica* | 59—*Lestes dorothea* | 60—*Megalestes major* | 61—*Calicnemia imitans* | 62—*Copera marginipes* | 63—*Drepanosticta carmichaeli.*

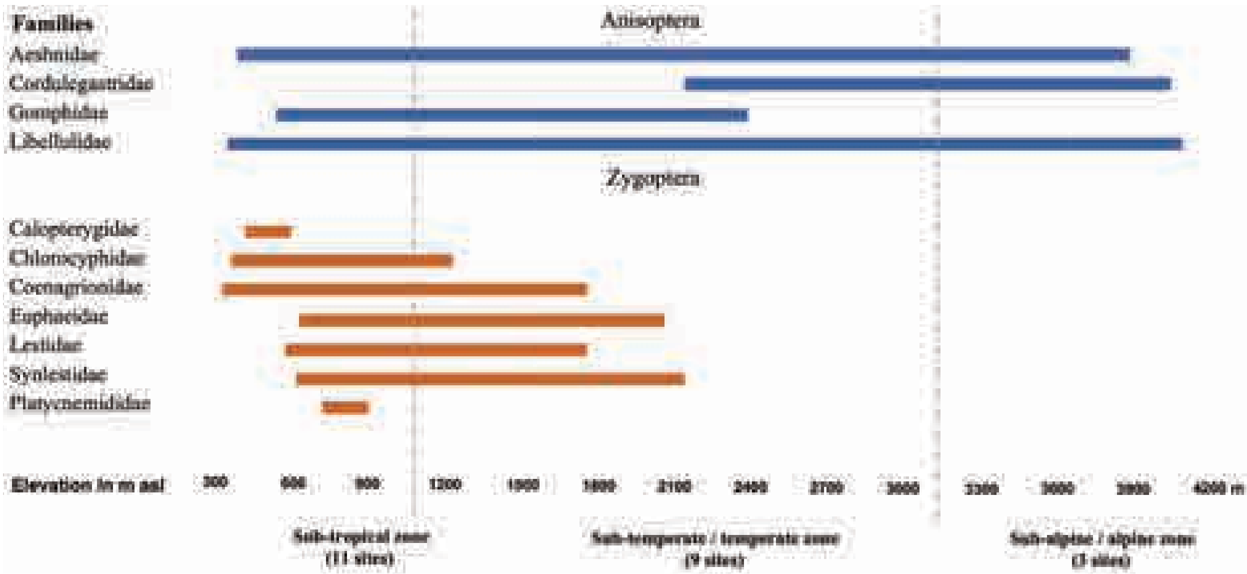

Figure 3. The elevational distribution of odonatan families in the study area. The blue bars represent Anisoptera, while the red bars represent Zygoptera. The dashed lines separate the three climatic zones, the subtropical (11 sites), subtemperate / temperate (9 sites) and alpine (3 sites).

Lestes dorothea, *Megalestes major*, and *Drepanosticta carmichaeli*. Among these *Sympetrum speciosum* and *Aristocypha trifasciata* reported by Singh (2022) are based on the media records of the first author already published in Odonata of India website (https://www.indianodonata.org/). The newly added Odonata fauna to the northwestern Himalaya include *Davidius davidii*, *Tramea transmarina*, and *Calicnemia imitans* (Table 2).

DISCUSSION

The Anisoptera families Libellulidae (29 species) and Aeshnidae & Gomphidae (5 species, each) accounted for nearly 62% of all species observed during the current sampling. Four families (Cordulegastridae, Calopterygidae, Lestidae, and Synlestidae) and 24 genera were monotypic indicating their restricted distribution in the region. Libellulidae and Gomphidae are well-distributed (Subramanian 2005) Anisoptera across the Indian subcontinent. The widespread dispersal and distribution may be attributed to the larger body size of species in these families (Dalzochio et al. 2011). The habitat heterogeneity and varying microclimatic regime sustain a high species richness and diversity (Cramer & Willig 2005; Storch et al. 2023) among different groups of animals. High species richness has been recorded from the small water bodies like rivulets, and streams as they create conducive microhabitats suitable for their survival as observed by Arunima & Nameer (2021), Chandran & Chandran (2021) Chandran et al. (2021), and Thakuria & Kalita (2021) as well. Key conditions for many species include shading around water bodies, specific vegetation structure for breeding and oviposition or nymphal microhabitat availability (Rantala et al. 2004; Cheri & Finn 2023). Subtropical ecosystems supported more odonates than temperate and alpine habitats, indicating a declining trend in species richness with increasing elevation as reported in other insect groups (Vetaas et al. 2019, Fontana et al. 2020; Dewan et al. 2022). No damselfly was found above 2,200 m in the current sampling effort (Figure 3). This does not, however, elude their presence in sub-alpine and alpine climate zones, as these landscapes were not visited as frequently as subtropical and temperate ones were.

Singh (2022) described 184 odonate species from north-western region of India, including 65 from the Union Territory of Jammu & Kashmir. During their explorations in selected localities of the Jammu division, Kumar et al. (2022) observed 32 odonate species, 25 of which are new to Jammu & Kashmir. This communication adds 24 more species to odonate fauna of Jammu & Kashmir, including three new to the northwestern Himalaya. This trend may be explained by the fact that the area has been less explored for Odonata. Recent studies conducted in the Himalaya reveals that Odonata fauna of the region is threatened due to habitat destruction, agricultural expansion, pesticides, tourism, urban and industrial pollution (Subramanian & Babu 2018; De et al. 2021) and this holds true for the study area. Aquatic ecosystems are spatially and temporally constrained (De et al. 2021), and the sites of current explorations are found in close proximity to human settlements, roads and highways making them vulnerable to management activities that threaten the existence of aquatic biodiversity including odonates. Most roadside ditches are being destroyed by road expansions, ponds are being encroached upon for land reclamation, and rivers and streams are being exposed to sand extraction and increased pollutant loads.

In terms of the occurrence data, 48 species belonged to occasional and rare categories. Arunima & Nameer (2021) in their observations recorded a moderately high number of occasional and rare species. Interestingly, all taxa found during the sampling figure in the least concern category of conservation (IUCN 2023) indicating a stable worldwide population. The study though preliminary with limited area coverage and ecological scope has unveiled vital information regarding the distribution of the observed odonate species in the heterogenous landscapes of northwestern Himalaya. Though the current observations on the Odonata do not necessarily provide a complete checklist for the region, they do add to the knowledge of the insect fauna of the Union Territory of Jammu & Kashmir. More systematic research on odonate assemblages and seasonality is needed to describe the ecology and biomonitoring of their habitats in the region as macroinvertebrates are standard bioindicators of freshwater ecosystems (Barbour et al. 1999).

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Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23557–23566
ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)
https://doi.org/10.11609/jott.7977.15.7.23557-23566
#7977 | Received 15 April 2022 | Final received 02 June 2023 | Finally accepted 28 June 2023

OPEN ACCESS

COMMUNICATION

Checklist of soil nematode diversity from Udupi District, Karnataka, India

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Abstract: Nematodes are plentiful in soil and may be found in practically every habitat. Around 25% of global biodiversity is considered to be supported by terrestrial ecosystem soils. There has been less research on nematode populations in Karnataka than there has been in other states. The scarcity of available literature provides up even more opportunities for studying these faunas in this region. As a result, the following investigation was conducted. The major goal of this research was to investigate the nematode diversity in the Udupi area. The collected nematodes were fixed, dehydrated, and displayed on a glass slide after isolation. Published keys were used to identify the species; there were 2,833 individual nematodes recovered. This collection contained 49 soil nematode species, which were classified into 34 genera and 20 families distributed over seven orders.

Keywords: Bacterial feeders, c-p values, fungal feeders, Mononchida, NEMAPLEX, Tylenchida.

Kannada: ದುಂಡುಹುಳಗಳು ಮಣ್ಣಿನಲ್ಲಿ ಹೇರಳವಾಗಿವೆ ಹಾಗೂ ಪ್ರತಿಯೊಂದು ಆವಾಸಸ್ಥಾನದಲ್ಲಿ ಅವುಗಳು ಕಂಡುಬರಬಹುದು. ಜಾಗತಿಕ ಜೀವವೈವಿಧ್ಯದ ಶೇಕಡಾ 25ರಷ್ಟು ಮಣ್ಣಿನಲ್ಲಿ ಜೀವಿಸುತ್ತವೆಂದು ಅಂದಾಜಿಸಲಾಗಿದೆ. ಕರ್ನಾಟಕದಲ್ಲಿ ದುಂಡುಹುಳಗಳು ಬಗ್ಗೆ ತೀರಾ ಕಡಿಮೆ ಸಂಶೋಧನೆ ನಡೆದಿರುವುದು, ಲಭ್ಯ ವೈಜ್ಞಾನಿಕ ದಾಖಲೆಗಳಿಂದ ಧೃಡಪಡುತ್ತದೆ. ಸಾಹಿತ್ಯದ ಲಭ್ಯತೆಯ ಕೊರತೆಯು ಈ ಪ್ರದೇಶದಲ್ಲಿ ಈ ಪ್ರಾಣಿಗಳನ್ನು ಅಧ್ಯಯನ ಮಾಡಲು ಇನ್ನಷ್ಟು ಅವಕಾಶಗಳನ್ನು ಒದಗಿಸುತ್ತದೆ. ಇದರ ಪರಿಣಾಮವಾಗಿ, ಈ ಸಂಶೋಧನೆಯನ್ನು ನಡೆಸಲಾಯಿತು. ಉಡುಪಿ ಪ್ರದೇಶದಲ್ಲಿ ಲಭ್ಯವಿರುವ ಮಣ್ಣಿನ ದುಂಡುಹುಳಗಳು ವೈವಿಧ್ಯತೆಯನ್ನು ಅಧ್ಯಯನ ಮಾಡುವುದು ಈ ಸಂಶೋಧನೆಯ ಪ್ರಮುಖ ಗುರಿಯಾಗಿದೆ. ಉಡುಪಿಯ ಬೇರೆ ಬೇರೆ ಭಾಗಗಳಿಂದ ಸಂಗ್ರಹಿಸಿದ ಮಣ್ಣಿನಿಂದ ದುಂಡುಹುಳಗಳನ್ನು ಬೇರ್ಪಡಿಸಿ ಅದನ್ನು ನಿರ್ಜಲೀಕರಿಸಿ ಗ್ಲಾಸ್ ಸ್ಲೈಡ್‌ನ ಮೇಲೆ ಅಂಟಿಸಿ ಆನಂತರ ಅವುಗಳ ಚಿತ್ರ ಹಾಗೂ ರೇಖಾಚಿತ್ರಗಳನ್ನು ತೆಗೆದುಕೊಳ್ಳಲಾಯಿತು. ನಂತರ ಈಗಾಗಲೇ ಲಭ್ಯವಿರುವ ಸಾಹಿತ್ಯವನ್ನು ಆಧರಿಸಿ ಅವುಗಳನ್ನು ಗುರುತಿಸಲಾಯಿತು. ಒಟ್ಟು 2833 ದುಂಡುಹುಳಗಳನ್ನು ಈ ರೂಪದಲ್ಲಿ ಸಂಗ್ರಹಿಸಿ ಅವುಗಳಲ್ಲಿ ಒಟ್ಟು 20 ಕುಟುಂಬಕ್ಕೆ ಸೇರಿದ 34 ತಳಿಯ 49 ಜಾತಿಯ ಮಣ್ಣಿನ ದುಂಡುಹುಳಗಳನ್ನು ಗುರುತಿಸಿ ಇಲ್ಲಿ ವರದಿ ಮಾಡಲಾಗಿದೆ.

Editor: Biplob K. Modak, Sidho Kanho Birsha University, Purulia, India.

Date of publication: 26 July 2023 (online & print)

Citation: Murthy, M.V.K. & A. Shwetha (2023). Checklist of soil nematode diversity from Udupi District, Karnataka, India. *Journal of Threatened Taxa* 15(7): 23557–23566. https://doi.org/10.11609/jott.7977.15.7.23557-23566

Funding: Self-funded.

Competing interests: The authors declare no competing interests.

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Author contributions: KM designed the study, conducted field work, data collection, data analysis and wrote the manuscript. SA supervised the research, designed the study, contributed in data analysis, and provided multiple revisions in the early stages of writing. Both authors read and approved the final manuscript.

Acknowledgements: Authors are grateful to Chairman, Department of Applied Zoology, Kuvempu University for extending all the help needed.

INTRODUCTION

Nematodes are ubiquitous in soil and occur in almost every type of ecosystem (Coleman et al. 2004). In terms of diversity and abundance, nematodes are one of the most diverse and abundant phyla in the animal kingdom. They have a high degree of genetic diversity and phenotypic plasticity, allowing them to colonize a wide variety of habitats. Nematodes are the most numerous multicellular animals that live in the soil, and feed and reproduce in the water film surrounding and within soil aggregates. Nematodes, which are comprised of over 30,000 described species, exist in almost all possible environment on the planet and account for more than 80% of metazoan taxonomic and functional diversity in soil (Nisa et al. 2021).

Terrestrial ecosystem soils are thought to sustain around 25% of global biodiversity. Although there are more than a million nematode species predicted, only about 30,000 have been discovered (Kiontke & Fitch 2013; Nisa et al. 2021). The greatest nematode abundance (309,000 individuals per kilogram of dry soil) was found around latitude 50^0, with an average of 27,600 individuals per kg of dry soil (Song et al. 2017). Nematodes are an essential component of the soil microbiota, aiding in the regulation of a wide range of ecosystem functions including mineral cycling, succession processes, and energy flow (Nisa et al. 2021).

In Karnataka, there have been comparatively fewer studies on nematode communities. The insufficiency of existing literature opens even greater possibilities for exploring these fauna in this area. Ravichandra & Krishnappa (2004) and Kantharaju et al. (2005) have studied the prevalence, distribution, pathogenicity, and control of economically important plant parasitic nematodes. It is reasonable to assume that investigations on nematodes other than commercially important species have not been conducted in the study region. As a consequence, the following investigation has been carried out. The primary purpose of this study was to explore the nematode diversity in the Udupi region.

MATERIALS AND METHODS

Study Area

Udupi is wedged between the Western Ghats on the east and the Arabian Sea on the west (Figure 1). Udupi district has an area of 3,880 km^2 and is situated at 13.33^0N & 74.74^0E at an average elevation of 27 m. The area of Udupi adjacent to the sea is plain with tiny hills, rice fields, coconut groves, and urban areas. Summers (March–May) can reach 38^0C, while winters (December–February) range 32–20 ^{0}C. The monsoon season lasts from June to September, with annual rainfall averaging over 4,000 mm (160 in) and strong winds (District Disaster Management Authority 2022).

Collection of soil samples

From each of Udupi's seven taluks, 25 soil and 25 sediment samples were collected. Soil cores were sampled using opportunistic sampling (Williams & Brown 2019). A soil auger or hand spade was used to collect soil and sediments. Sampling was done at a depth of 10 to 15 cm in the early hours of the day. Five to six cores of soil around the plant roots were excavated, and roughly 1 kg of soil was collected and put into zip lock polythene bags, which were then immediately moved to a chiller with a temperature of 4^0C, and carried until further processing (Ravichandra 2014).

Isolation of nematodes from soil

Nematodes were isolated employing Cobb's sieving and decanting technique. The murky filtrate was then subjected to Bearman's Funnel technique for isolation (Sikora et al. 2018).

Killing, processing, and fixing the nematodes

The nematode suspension thus obtained was placed in a test tube for 20–30 minutes to allow the nematodes to settle to the bottom. The bulk of the water was gently emptied from the test tube using a dropper and killed suddenly by plunging it in hot 4% formalin (heated to 60° C). Killed nematodes were fixed in 5 parts of glycerine and 95 parts of alcohol fixative and allowed for slow dehydration in a desiccator with calcium chloride as a desiccant for about three weeks (Ravichandra 2014).

The fixed nematodes were then carefully extracted, and permanent slides were made by employing the wax ring technique with a drop of pure anhydrous glycerine. Toup-view micrometry software was used to make measurements, and de man's indices (de Man 1884) were used to make measurements (Sikora et al. 2018). Species were identified following keys available in Siddiqui (2000), Ahmad & Jairajpuri (2010), Bohra (2011), and the NEMAPLEX website (Nemaplex 2022). Each individual was assigned to respective trophic group according to Yeates et al. (1993) and various feeding habits according to Bongers & Bongers (1998).

Figure 1. Map of the study area.

RESULTS

The total number of individual nematodes isolated from the soils collected from the research area was 2833. This comprised of 49 species of soil nematodes belonging to 34 genera and 20 families distributed among seven orders. Order Tylenchida was the most dominant order represented by 27 species (57%) followed by the Dorylaimida with 11 species (23%), Aphelenchida with four species (8%), Mononchida with three species (6%), Rhabditida with two (4%), Araeolaimida (2%), and Monhysterida (2%) were represented by a species each (Figure 2). Family Qudsianematidae and Tylenchidae were the families comprising the highest number of species (Figure 3). The detailed family-wise species representation is displayed in Table 1. Photographs of few selected nematodes are given in Image 1–34.

Yeats et al. (1993) identified eight distinct types of nematode feeding. The feeding categories have also been attributed to the species inventory of the present study. The species that belong to feeding type 1 (plant feeders) are the most prevalent community, with 24 species representing the category, nine species belong to feeding group 5 (predators), six to feeding type 8

Table 1. Names of documented species (with feeding type) and their family. (With C-p values and feeding habit). All names are after Bohra (2011)

	Name of the species (under various families)	C-p Value	Feeding habit
	Family 1: Anguinidae		
1.	*Ditylenchus clarus* Thorne and Malek, 1968	2	Fungal-feeding
	Family 2: Aphelenchoididae		
2.	*Aphelenchoides asterocaudatus* Das, 1960	2	Plant-feeding
3.	*Aphelenchoides longiurus* Das, 1960	2	Plant-feeding
4.	*Aphelenchoides besseyi* Christie, 1942	2	Plant-feeding
5.	*Aphelenchoides bicaudatus* (Imamura, 1931) Filipjev and Stekh., 1941)	2	Plant-feeding
	Family 3: Cephalobidae		
6.	*Zeldia puntata* (Thorne, 1925) Thorne, 1937	2	Bacterial-feeding
7.	*Cephalobus bodenheimeri* (Stainer, 1936) Andrassy, 1984	2	Bacterial-feeding
	Family 4: Dorylaimidae		
8.	*Mesodorylaimus mesonyctius*	4	Omnivore
9.	*Dorylaimis stagnalis* Dujardin, 1835	4	Omnivore
10.	*Mesodorylaimus margeritus* Basson and Heyns, 1974	4	Omnivore
11.	*Laimydorus serpentines* (Thorne and Swanger, 1936) Siddiqi, 1969	4	Omnivore
	Family 5: Hoplolaimidae		
12.	*Helicotylenchus martini* Sher, 1960	3	Plant-feeding
13.	*Hlelicotylenchus indicus* Siddiqi and Husain, 1964	3	Plant-feeding
14.	*Helicotylenchus digitatus* Siddiqi and Husain, 1964	3	Plant-feeding
	Family 6: Iotonchidae		
15.	*Iotonchus trichuris* (Cobb, 1917) Mulvey, 1963	4	Predators
	Family 7: Longidoridae		
16.	*Longidorus proximus* Sturhan and Agro, 1983	5	Plant-feeding
17.	*Longidours minrus* Khan et al., 1972	5	Plant-feeding
18.	*Longidorus elongatus* (de Man, 1876) Micoletzky, 1922	5	Plant-feeding
19.	*Paralongidorus sp*	5	Plant-feeding
	Family 8: Meloidogynidae		
20.	*Meloidogyne javanica* (Treub, 1885) Chitwood, 1949	5	Plant-feeding
21.	*Meloidogyne incognita* (Kofoid and White, 1919) Chitwood, 1949	3	Plant-feeding
22.	*Heterodera cajani* Koshi, 1967	3	Plant-feeding
23.	*Heterodera zeae* Koshy, Swarup and Sethi, 1971	3	Plant-feeding
	Family 9: Monhysteridae		
24.	*Monhystera* spp.	2	Bacterial-feeding
	Family 10: Mononchidae		
25.	*Mononchus* spp.	4	Specialist Predators
	Family 11: Mylonchulidae		
26.	*Mylonchulus minor* (Cobb, 1893) Andrassy, 1958	4	Specialist Predators
	Family 12: Nordiidae		
27.	*Kochinema sectum* Siddiqi, 1966	4	Generalist predators
	Family 13: Nygolaimidae		
28.	*Nygolaimus anneckei* Heyns, 1969	5	Generalist predators
	Family 14: Paratylenchidae		
29.	*Paratylenchus curvitatus* Van der Linde, 1938	2	Plant-feeding
30.	*Paratylenchus nainianus* Edward and Misra, 1963	2	Plant-feeding
	Family 15: Plectidae		
31.	*Plectus parvus* Bastian, 1865	2	Bacterial-feeding
	Family 16: Pratylenchidae		
32.	*Pratylenchus coffeae* (Zimmerman, 1898) Filipjev and Stekhoven, 1941	3	Plant-feeding
33.	*Pratylenchus thornei* Sher and Allen, 1953	3	Plant-feeding
	Family 17: Qudsianematidae		
34.	*Eudorylaimus centrocercus* (De Man, 1880) Andrassy, 1959	4	Generalist predators
35.	*Eudorylaimus longicardiu,* Thorne, 1974	4	Generalist predators
36.	*Discolaimus rotundicaudatus,* Khan and Laha, 1982	4	Generalist predators
37.	*Moshajia cultristyla* Siddiqi, 1982	4	Generalist predators
38.	*Discolaimus agricolus* Sauer and Annells, 1986	4	Generalist predators
39.	*Discolaimus major* Thorne, 1939	4	Generalist predators
	Family 18: Telotylenchidae		
40.	*Tylenchorhynchus zeae* Sethi and Swarup, 1968	3	Plant-feeding
41.	*Tylenchorhynchus clarus* Allen, 1955	3	Plant-feeding
42.	*Qunisulcius capitatus*	3	Plant-feeding
	Family 19: Tylenchidae		
43.	*Tylenchus magnus* Khurana and Gupta, 1988	2	Plant-feeding
44.	*Aglenchus agricola (de Man, 1884) Meyl, 1961*	2	Plant-feeding
45.	*Filenchus filifornis* (Brzeski, 1963) Lownsbery and Lownsbery, 1985	2	Plant-feeding
46.	*Sakia alii* Suryawanshi, 1971	2	Plant-feeding
47.	*Boleodorus brevistylus* Khera, 1970	2	Plant-feeding
48.	*Basiria graminophila* Siddiqi, 1959	2	Plant-feeding
	Family 20: Xiphinematidae		
49.	*Xiphinema americanum* Cobb, 1913	5	Plant-feeding

1–5—colonizers – persisters | c-p-value—structural guild: 1—enrichment opportunists | 2—basal fauna | 3—early successional opportunists | 4—intermediate succession and disturbance sensitivity | 5—long-lived intolerant species. Allotments follow Bongers & Bongers (1998).

(omnivores), six to feeding type 2 (hyphal feeding) and four to feeding type 3, which includes bacterial feeders. A further inspection of the pooled data reveals that plant-feeding taxa form a significant trophic community in this region, with omnivore and fungal feeders having relatively little representation. Herbivore nematode fauna is relatively higher when compared to the other groups probably due to the restriction of sampling sites to the areas with lush vegetation. Allocation of documented taxa to various trophic guilds following Yeats et al. (1993) indicated that throughout the documented nematode families, there are nine plant-feeding, six predatory, three bacterial feeders, one omnivore, and a fungal-feeding nematode family.

Figure 2. Percent representation of different nematode orders.

C-p values (Colonizer-Persister) were allocated to each documented family following Bongers & Bongers (1998) (Table 3). Soil nematodes were classified into one of five colonizer-persister groups which range between extreme r- to extreme k-strategists. “Colonizer” nematodes at the lower end of the scale of the c-p scale are thought to be enrichment opportunists and so suggest resource availability; “persister” nematodes at the high end of the scale imply system stability, food web complexity, and connectance. C-p value range from 1 to 5 where the classification is mainly based on lifespan (Increasers with the scale), gonad to body volume (Increasers with the scale), sensitivity to soil perturbances which also increases with the scale and hence indicate the health of the soil.

DISCUSSION

This is a preliminary (possibly the first) study that focuses on the overall diversity of soil nematode communities in the Udupi region. We want to continue

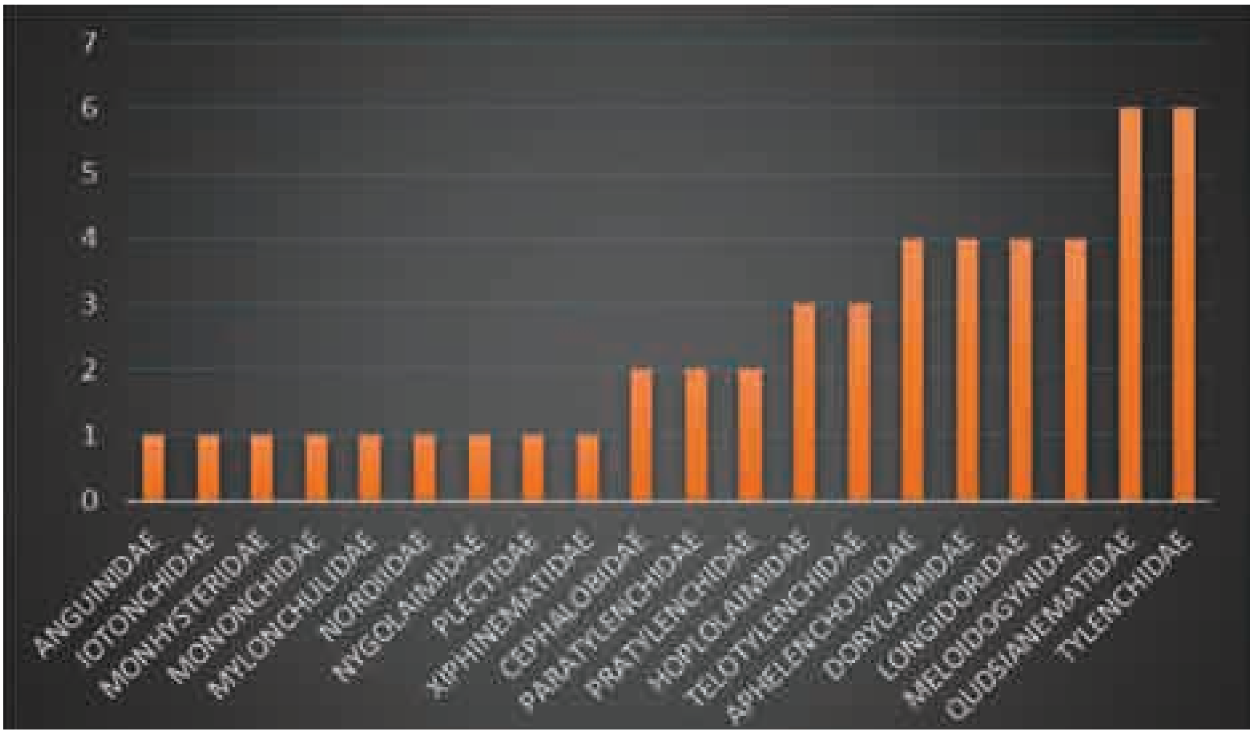

Figure 3. Number of species representing different nematode families.

Image 1–9. 1—*Ditylenchus clarus* | 2—*Aphelenchoides asterocaudatus* | 3—*Aphelenchoides longiurus* | 4—*Aphelenchoides besseyi* | 5—*Dorylaimis stagnalis* | 6—*Laimydorus serpentine* | 7—*Helicotylenchus martini* | 8—*Helicotylenchus digitatus* | 9—*Iotonchus trichuris*. Scale: 1,2,3 & 9—10 µm | 4–8—100 µm. © Keshava Murthy M V.

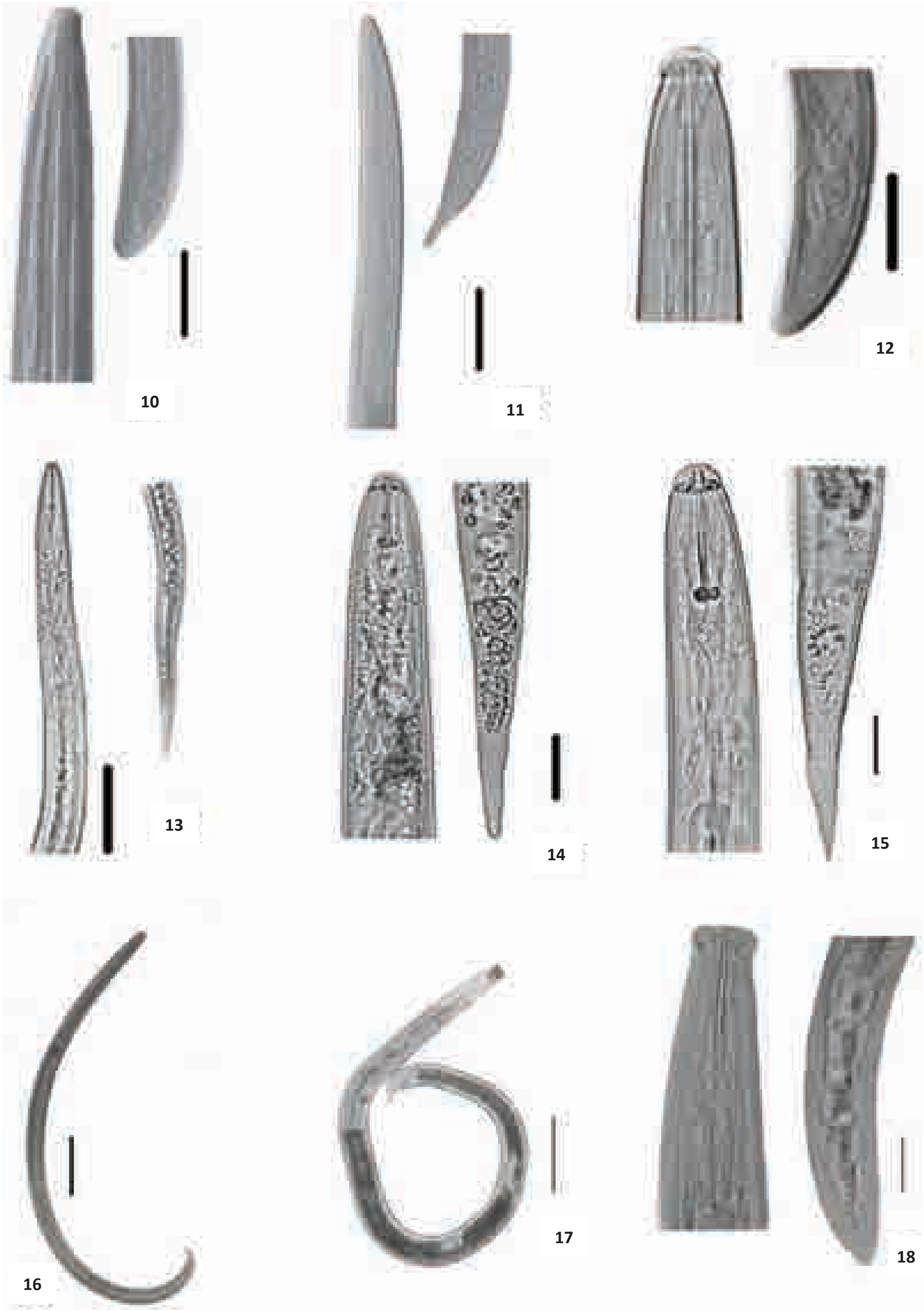

Image 10–18. **10—*Longidorus proximus* | 11—*Longidours minrus* | 12—*Paralongidorus* sp. | 13—*Meloidogyne javanica* | 14—*Meloidogyne incognita* | 15—*Heterodera zeae* |16—*Mononchus* sp. | 17—*Mylonchulus minor* | 18—*Kochinema sectum*. Scale: 10–15 & 18—10 µm | 16–17—100 µm. © Keshava Murthy M V.**

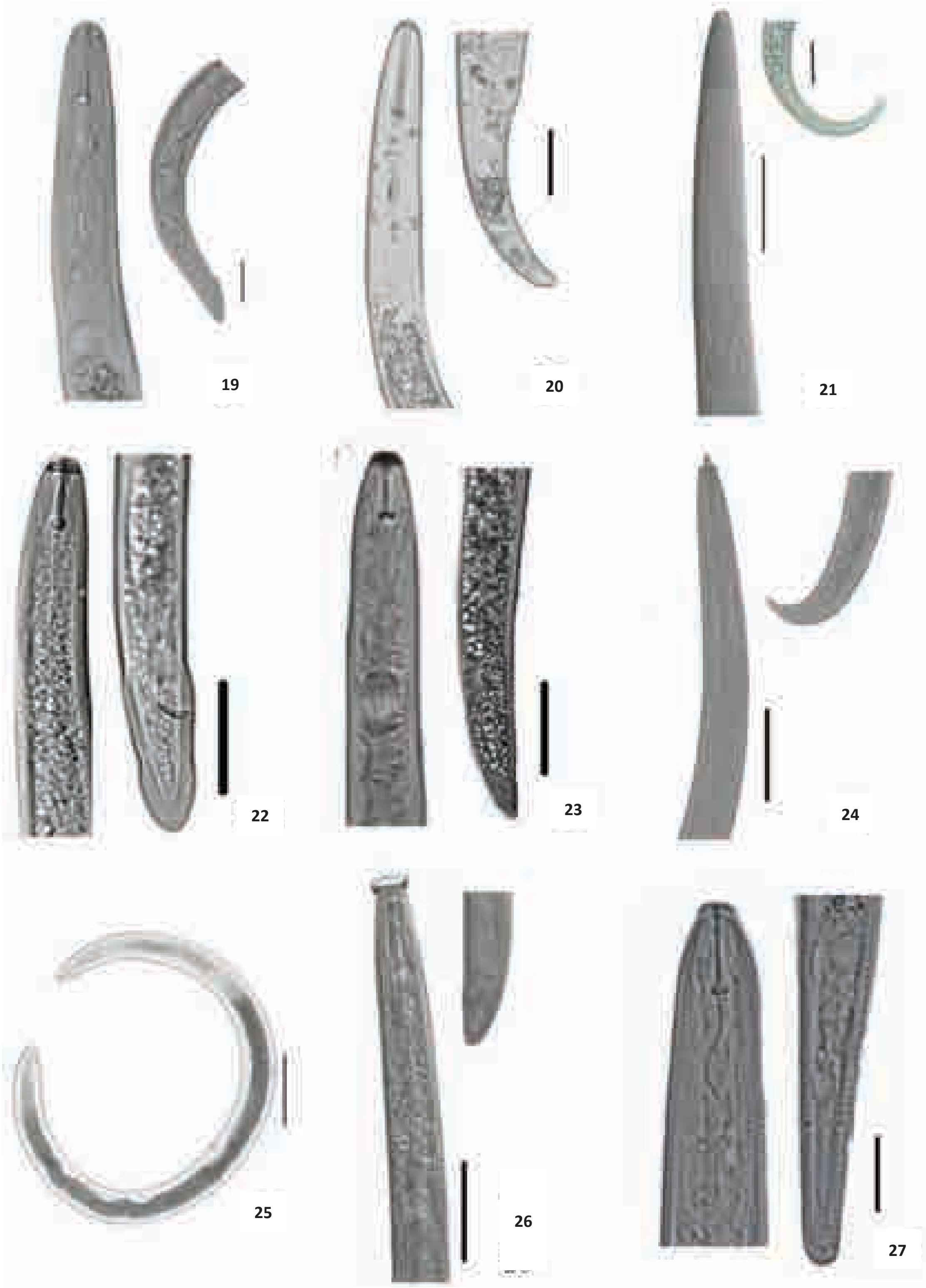

Image 19–27. 19—*Paratylenchus curvitatus* | 20—*Paratylenchus nainianus* | 21—*Plectus parvus* | 22—*Pratylenchus coffeae* | 23—*Pratylenchus thornei* | 24—*Eudorylaimus centrocercus* | 25—*Eudorylaimus longicardius* | 26—*Discolaimus agricolus* | 27—*Tylenchorhynchus clarus*. Scale: 19–24 & 26–27—10 µm | 25—100 µm. © Keshava Murthy M V.

Image 28–34. 28—*Tylenchus magnus* | 29—*Aglenchus agricola* | 30—*Filenchus filifornis* | 31—*Boleodorus brevistylus* | 32—*Basiria graminophila* | 33—*Xiphinema amerincanum* | 34—*Tylenchorhynchus zeae*. Scale: 28–30 & 33–34—10 µm | 31—50 µm | 32—75 µm. © Keshava Murthy M V.

the research, taking into account many soil parameters that influence nematode bioecology, to uncover the likely drivers of nematode assemblages in the soil of Udupi district. Nematodes are good models of soil health indicators since they are widespread and distributed over a variety of feeding behaviors and trophic guilds (Kergunteuil et al. 2016). It's astounding that microbial biogeography still lacks a map, given that the great majority of biodiversity is found in microscopic taxa rather than macroscopic taxa. Also, considering that microscopic species play critical roles in ecosystem functioning via decomposition and nutrient mineralization processes, it is surprising that we still don't know much about patterns of nematode diversity and nematode assemblages in soil ecosystems (Porazinska et al. 2012). More comprehensive studies on nematode populations in Udupi might yield exciting results that help us to monitor soil quality and, if required, to design and implement mitigation strategies.

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Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23567–23574
ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)
https://doi.org/10.11609/jott.8272.15.7.23567-23574
#8272 | Received 18 November 2022 | Final received 20 May 2023 | Finally accepted 30 May 2023

COMMUNICATION

Checklist of the genus *Dendrobium* Sw. (Orchidaceae) in Manipur, India

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Abstract: An enumeration of the genus *Dendrobium* Sw. (Orchidaceae) in Manipur, India was attempted. Literature review revealed the occurrence of 67 species under the genus from the state. The study was carried out through repeated field explorations in different natural forests of Manipur since 2012. Out of the total taxa reported from the state, only 42 species could be traced in the field. All these taxa are presented here with their flowering phenology and places of occurrence. Twenty-five species were untraceable in the field; however, nine species out of these 25 are represented by authentic herbarium specimens and supported by published documents. The remaining 16 species were reported only in literature, but no live plants or herbarium specimens were found. Out of the 42 species inventoried from the field, many taxa are under threat owing to habitat loss due to felling of trees, deforestation, and 'jhum' cultivation. So, there is an urgent need of conservation of those species through in situ and ex situ means for their sustenance. A photographic plate of some taxa is provided here which might be useful for their easy identification in the field and for taking care of their conservation. Large-scale uses of these species as cut flowers and indoor & outdoor plants may be practised through micropropagation and cultivation in nurseries which may help in revenue generation for the state.

Keywords: conservation, inventorization, northeastern India, orchid, threats.

Editor: Pankaj Kumar, Department of Plant and Soil Science, Texas Tech University, Lubbock, Texas, USA. **Date of publication:** 26 July 2023 (online & print)

Citation: Sharma, H.B. & D. Bhattacharyya (2023). Checklist of the genus *Dendrobium* Sw. (Orchidaceae) in Manipur, India. *Journal of Threatened Taxa* 15(7): 23567–23574. https://doi.org/10.11609/jott.8272.15.7.23567-23574

Funding: DST, Government of India, New Delhi.

Competing interests: The authors declare no competing interests.

Author details: Dr. Hidangmayum Bishwajit Sharma did his research on the morpho-taxonomic studies on the genus *Dendrobium* in Manipur and was awarded PhD under the supervision of Dr. Debjyoti Bhattacharyya. Presently, he is serving as a Government employee under the State Government of Manipur. Dr. Debjyoti Bhattacharyya is an associate professor in the Department of Life Science & Bioinformatics, Assam University, Silchar. He acted as the supervisor of Dr. H. Bishwajit Sharma.

Author contributions: First author (HBS) collected the specimens from the field, worked out, identified, prepared the specimens for the herbarium and drafted the manuscript. Corresponding author (DB) supervised the work, checked the manuscript and communicated it to the journal.

Acknowledgements: Authors are thankful to the head, Department of Life Science & Bioinformatics, Assam University, Silchar for facilities. First author is grateful to Mr. H. Kipgen, president (FEEDS & KVK, Hengbung) and the DST, Government of India, New Delhi for providing facilities and financial support during the study.

INTRODUCTION

The members of Orchidaceae Juss. are well known in the world for their beautiful and ineffable flowers of different colours, hues, and forms. Taxonomically, Orchidaceae is one of the most diversified and evolved families of flowering plants (Kumar & Manilal 1994). There is a vast range of diversity in the shape, colour, and size of orchid flowers; yet they are the same in their basic form (Pradhan 2005). Theophrastus (370–285 B.C.) named the group of bizarre plants as 'Orchids' finding resemblance of below-ground paired tubers with male testicles.

Dendrobium Sw. is the second largest genus of the family Orchidaceae after *Bulbophyllum* Thouars. It was established by Olavo (Peter) Swartz in 1799. The name was derived from the Greek words 'dendron' (tree) and 'bios' (life) which means 'one who lives on trees', or essentially 'epiphytes'. The genus is divided into 14 sections, viz.: *Bolbidium*, *Callista*, *Dendrobium*, *Breviflores*, *Formosae*, *Stachyobium*, *Pedilonum*, *Distichophyllum*, *Rhopalanthe*, *Aporum*, *Oxystophyllum*, *Strongyle*, *Grastidium,* and *Conostalix* (Seidenfaden 1985). The genus is sympodial with varying length of pseudobulbs. Most of the species are generally epiphytic or occasionally lithophytic in nature. They have adapted themselves to a wide variety of habitats starting from high altitudes to lowland tropical forests. They remain dormant during winter but quickly grow in summer. In spring, occasionally in autumn, dormant buds come out from the base of the pseudobulb followed by fast growth of new roots. Most of the Dendrobiums flower during the pre-monsoon season (March–May) and a few species blossom in the post-monsoon period. Capsules are produced late in the dry season (August–December).

The genus *Dendrobium* comprises about 3,160 species (Govaerts et al. 2022) with high morphological diversity and is mainly distributed in Sino-Himalayan regions with further extension up to Australia, New Zealand, and Pacific Islands (Wood 2006). In India, the genus is represented by c. 117 species (Rao 2022), of which, about 88 species are found in northeastern India (Rao 2018).

A checklist of the genus *Dendrobium* in Manipur was prepared from different literature (Mukerjee 1953; Deb 1956, 1957, 1960, 1961; Phukan 1996; Chauhan 2001; Kumar & Kumar 2005; Nanda et al. 2012, 2013, 2014; Kishor et al. 2013; Meitei et al. 2014; Khuraijam et al. 2016; Deori et al. 2019), which documents the occurrence of 67 species in the state.

MATERIALS AND METHODS

Study area

The state of Manipur, a part of Indo-Burma Biodiversity Hotspot, is bounded by Nagaland in the north, Mizoram in the south, Assam in the west, and Myanmar (Burma) in the east as well as in the south (Image 1). The state lies between the coordinates $23^0$83'–$25^0$68' N and $93^0$03'–$94^0$78' E. The total geographical area covered by the state is 22,327 km^2. The total forest cover of Manipur is 17,219 km^2, which is 77.12% of the total geographical area of the state. The state lies in a unique geographical position between the virtual meeting point of India and southeastern Asia (Singh 2014) with a total boundary of 854 km length. The altitude varies 50–3,000 m.

Based on the topography, structure, geology, the location's relief, and other geographical conditions, Manipur can be divided into two major natural physiographic divisions, viz.: (i) The Manipur hills and mountains and (ii) The Manipur valleys. Five major types of forests are prevalent in the state. These are: 1. Subtropical semi-evergreen forests, 2. Subtropical deciduous forests, 3. Montane wet temperate forests, 4. Subtropical pine forests, and 5. Subtropical dry temperate forests (Singh 2014).

Field survey and data collection

For the present study, several field explorations were conducted in different places of Manipur since 2012 (Table 1). Field surveys were made covering all seasons, although pre-monsoon and post-monsoon are the best collection seasons for the orchids especially for the genus *Dendrobium*. Locations were noted with their altitudes and geographical coordinates. A total of 58 sites were visited (Table 1). Surveys were conducted for at least 3–5 days at each location.

Identification of species

After collection, identification of species was done using standard methods of morpho-taxonomic studies. Flowers were dissected and critically studied under Stereo Zoom dissecting microscope (Olympus SZ61). Species without flowering were collected in a vegetative stage and grown in the nursery of the orchidarium of Centre for Orchid Gene Conservation of Eastern Himalayan Region (COGCEHR), Hengbung, Kangpokpi district, Manipur. These species were studied after they bloomed in the orchidarium. Morphological attributes were noted and identification of the species for all taxa were made using primary and secondary sources of

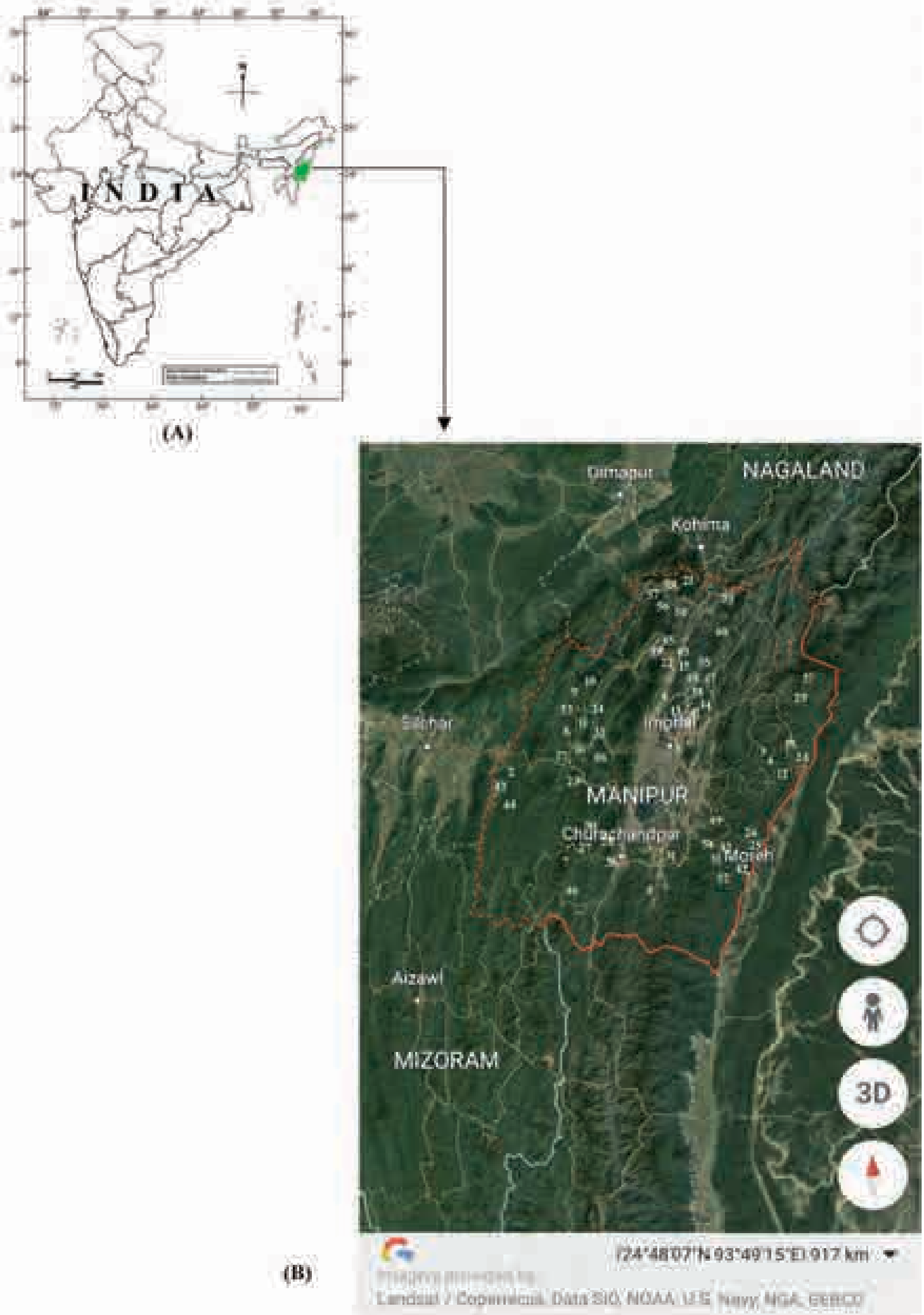

Image 1. A—Map of India showing study area | B—A Google Earth map (Data SIO, NOAA, U.S.Navy, NGA, GEBCO © 2017 Google) showing specific collection sites in the study area Manipur (Image taken on 08 April 2023 at 1405 h). (Corresponding Table 1 is referred for locality names).

Table 1. List of localities with their elevational range. Locality number corresponds to numbers in Image 1. (partly reproduced from Sharma, 2019).

	Sites of occurrence	Elevation range (in m)
1.	Baruni Hills	1100–1400
2.	Bidyanagar	50–60
3.	Bungpa Khullen	1100–1400
4.	Bungpa Khunou	1100–1400
5.	Chakpikarong	700–900
6.	Changoubung Nepali Khul	1250–1750
7.	Chiru	1000–1300
8.	Chiulon	1000–1328
9.	Dailong Cemetery	950–1260
10.	Dailong Rangan	900–1200
11.	Duigailong	1050–1350
12.	Grihang	1000–1300
13.	Haipi	1100–1500
14.	Hengbung	950– 1168
15.	Injolum	970–1200
16.	Kahulong	950–1260
17.	Kaikao	700–1000
18.	Kamjong	1230–1500
19.	Kapung Hill	1540–1752
20.	Keithelmanbi	900–1100
21.	Kenelu	1741–1832
22.	Khajinglok	1200–1450
23.	Khongsang	400–700
24.	Kongkan	1000–1300
25.	Kwatha	400–540
26.	Kwatha khulen	450–590
27.	Laimaton	1100–1600
28.	Lamdan	900–1300
29.	Langli	1500–2000
30.	Leimatak	450–600
31.	Litan Village	800–1010
32.	Lokchao	400–500
33.	Longchum	500–800
34.	Longku	1000–1250
35.	Lungdi Hill	1580–1942
36.	Majuron	900–1400
37.	Maku	1200–1500
38.	Malingli	1450–1600
39.	Mao	1200–1798
40.	Maram Khunuo	900–1345
41.	Mayangkhang	900–1150
42.	Moreh	400–550
43.	New Alipur	50–70
44.	Ningshingkhul	50–70
45.	Oklong	1350–1760
46.	Rangkhung	800–1100
47.	Sadim Pukhri	1300–1570
48.	Sadim Village	1250–1450
49.	Saivom Village	1300–1450
50.	Sangkungmai	990–1779
51.	Sinam Village	1350–1550
52.	Songpiyang	420–500
53.	Tamenglong	1000–1450
54.	Tengnoupal	1050–1400
55.	Tringalung	1420–1512
56.	Willong	955–1756
57.	Willong Khunou	850–1028
58.	Yangkhulen	970–1800

information, i.e., flora, monographs, articles, and books (Hooker 1890; Seidenfaden 1985; Kumar & Kumar 2005; Wood 2006; Lucksom 2007). Identity of the species was further confirmed by matching the specimens with the types and authentic herbarium specimens housed in the Central National Herbarium, Botanical Survey of India, Howrah (CAL) and Eastern Regional Centre, Botanical Survey of India, Shillong (ASSAM). Online databases, viz., The International Plant Names Index (IPNI 2022), Plants of the World Online (POWO 2022), Tropicos (2022), and The World Flora Online (WFO 2023) were consulted for updated nomenclature. Global Biodiversity Information Facility (GBIF 2023) was also browsed for digital images of species. Jain & Rao (1977) and Singh & Subramaniam (2008) were followed for preparation of herbarium sheets. Specimens were deposited in the Central Herbarium of Assam University, Silchar (AUSCH), Assam.

RESULTS

Out of the 14 sections of the genus *Dendrobium* (Seidenfaden 1985), species occurring in the state of Manipur are represented by eight sections (Table 2). In the field, the authors could locate only 42 species out of the 67 species recorded earlier from the state. Among these, 25 species could not be found in the wild, nine species are represented by herbarium specimens and

Table 2. List of species of *Dendrobium* located in their natural wild habitats in Manipur with flowering phenology, occurrence, and exsicata.

	Sections	Scientific name	Phenology	Voucher specimen(s)	Occurrence*
1.	Callista (Lour.) Schltr.	*Dendrobium chrysotoxum* Lindl.	April–June	H. Bishwajit Sharma 001,041,056	25, 47, 50
2.		*Dendrobium densiflorum* Lindl.	April–June	H. Bishwajit Sharma 002,010,011,080	57, 8, 17, 37
3.		*Dendrobium jenkinsii* Wall. ex Lindl.	March–June	H. Bishwajit Sharma 003,051,052,057	56, 26, 2, 43
4.		*Dendrobium lindleyi* Steud.	March–April	H. Bishwajit Sharma 012	31
5.	Formosae (Benth. & Hook.f.) Hook.f.	*Dendrobium draconis* Rchb.f.	June–July	H. Bishwajit Sharma 071	51
6.		*Dendrobium formosum* Roxb. ex Lindl.	August–September	H. Bishwajit Sharma 023	16
7.		*Dendrobium infundibulum* Lindl.	March–April	H. Bishwajit Sharma 072	54
8.		*Dendrobium longicornu* Lindl.	August–September	H. Bishwajit Sharma 008	58
9.		*Dendrobium tamenglongense* R.Kishor, Y.N.Devi, H.B.Sharma, J.Tongbram & S.P.Vij	July–September	Nanda 00510	16
10.		*Dendrobium williamsonii* Day & Rchb.f.	March–April	H. Bishwajit Sharma 045,049,088	47, 6, 24
11.	Breviflores Hook.f.	*Dendrobium bicameratum* Lindl.	July–August	H. Bishwajit Sharma 022,040,048	27, 14, 23
12.		*Dendrobium stuposum* Lindl.	June–July	H. Bishwajit Sharma 087	4
13.	Dendrobium	*Dendrobium amoenum* Wall ex Lindl.	May–June	H. Bishwajit Sharma 081	18
14.		*Dendrobium aphyllum* (Roxb.) C.E.C.Fisch.	March–April	H. Bishwajit Sharma 013,014,047,076	6, 53,11,1
15.		*Dendrobium bensoniae* Rchb.f.	June–July	H. Bishwajit Sharma 058	25
16.		*Dendrobium brymerianum* Rchf.f.	July–August	H. Bishwajit Sharma 027	45
17.		*Dendrobium capillipes* Rchb.f.	April–May	H. Bishwajit Sharma 034,035	36, 7
18.		*Dendrobium chrysanthum* Wall. ex Lindl.	August–September	H. Bishwajit Sharma 077,082	1, 12
19.		*Dendrobium crepidatum* Lindl. & Paxton.	April–May	H. Bishwajit Sharma 015,028,036,083	22, 3, 7, 19
20.		*Dendrobium crystallinum* Rchb.f.	April–May.	H. Bishwajit Sharma 059,060	42, 52
21.		*Dendrobium denneanum* Kerr.	May–June	H. Bishwajit Sharma 061,062,063	25, 49, 32
22.		*Dendrobium devonianum* Paxton.	April–May	H. Bishwajit Sharma 016,029,042	47, 46, 40
23.		*Dendrobium falconeri* Hook.	May–June	H. Bishwajit Sharma 030,043,064	55, 48, 52
24.		*Dendrobium fimbriatum* Hook.	April–May	H. Bishwajit Sharma 004,005,017	35, 33, 21
25.		*Dendrobium gibsonii* Paxton.	July–August	H. Bishwajit Sharma 018,084	34, 3
26.		*Dendrobium heterocarpum* Wall. ex Lindl.	April–May	H. Bishwajit Sharma 065,066,067	26, 54, 49
27.		*Dendrobium lituiflorum* Lindl.	April–May	H. Bishwajit Sharma 006,053,085	56, 4, 44
28.		*Dendrobium moschatum* (Buch.-Ham.) Sw.	June–July	H. Bishwajit Sharma 068,069	25, 42
29.		*Dendrobium nobile* Lindl.	March–April	H. Bishwajit Sharma 031,078,090,091	1, 13, 20, 39
30.		*Dendrobium ochreatum* Lindl.	April–May	H. Bishwajit Sharma 019,032,037,054	9, 2, 28, 39
31.		*Dendrobium parishii* Rchb.f.	May–June	H. Bishwajit Sharma 086	18
32.		*Dendrobium polyanthum* Lindl.	March–April	H. Bishwajit Sharma 007,020,038,044,079	57, 38, 1, 30, 17
33.		Dendrobium *pulchellum* Roxb. ex Lindl.	May–June	H. Bishwajit Sharma 070	26
34.		*Dendrobium transparens* Wall. ex Lindl.	May –June	H. Bishwajit Sharma 021,055	2, 5
35.		*Dendrobium wardiamum* R.Warner	April–May	H. Bishwajit Sharma 039	27
36.	Stachyobium Lindl.	*Dendrobium denudans* D.Don.	September–October	H. Bishwajit Sharma 009,024	58, 16
37.		*Dendrobium eriiflorum* Griff.	September–October	H. Bishwajit Sharma 073,089	29, 51
38.		*Dendrobium sinominutiflorum* S.C.Chen, J.J.Wood & H.P.Wood.	September–October	H. Bishwajit Sharma 033	45
39.	Pedilonum (Bl.) Lindl.	*Dendrobium cumulatum* Lindl.	July–August	H. Bishwajit Sharma 074	25
40.		*Dendrobium parcum* Rchb.f.	March–April	H. Bishwajit Sharma 025,050	10, 41
41.	Aporum (Bl.) Lindl.	*Dendrobium spatella* Rchb.f.	August–September	H. Bishwajit Sharma 026,075	25, 53
42.	Grastidium (Bl.) J.J.Smith	*Dendrobium salaccense* (Blume) Lindl.	March–April	H. Bishwajit Sharma 046	15

*For locality identification, refer to Table 1 and Image 1

Table 3. *Dendrobium* species represented only by herbarium specimens

	Scientific name	Locality	Voucher specimen
1.	*Dendrobium bellatulum* Rolfe	Senapati Hills, Senapati district, Manipur	A. A. Mao & R. Gogoi 111162 (ASSAM !)
2.	*Dendrobium cariniferum* Rchb.f.	Sirohi forests, Ukhrul District, Manipur	G. Watt 6500 (CAL !).
3.	*Dendrobium delacouri* Guillaumin	Yangoupokpi Lokchao Wildlife Sanctuary, Chandel district, Manipur	N.N. Rabha & L.R. Meitei 131115 (ASSAM !)
4.	*Dendrobium dickasonii* L.O.Williams	1500 m, Manipur	U.C.Pradhan 27 (K, Digital Image !)
5.	*Dendrobium kentrophyllum* Hook.f.	Sangaithel, Senapati district, Manipur	J.S. Khuraijam 302107 (LWG, Photo !)
6.	*Dendrobium moniliforme* (L.) Sw.	Phungum, Manipur	S. K. Mukerjee-2855 (CAL !)
7.	*Dendrobium monticola* P.F.Hunt & Summerh.	Karong, 3500 ft., Manipur	Thakur Rup Chand 3730 (MICH, Digital Image !)
8.	*Dendrobium porphyrochilum* Lindl.	Ukhrul, Ukhrul district, Manipur	S. K. Mukerjee 3420 (CAL !)
9.	*Dendrobium wattii* (Hook.f.) Rchb.f.	*s.l., s.d*, Manipur	G. Watt 5944 (CAL !)

authenticated by published documents (Table 3) and 16 species were mentioned in literature without any representative specimens from Manipur (Table 4).

All the collected species by the authors are presented here with their scientific and vernacular names, phenology, distribution in the state (Table 2). Photographs of the species which are very rare in the field are provided to ease the identification of the taxa.

Among the 42 collected species from the state under eight sections, the dominant section was *Dendrobium* which was represented by 23 species. It was followed by the section *Formosae* (6 spp.), *Callista* (4 spp.), *Stachyobium* (3 spp.), *Breviflores* (2 spp.), and *Pedilonum* (2 spp.). Sections *Aporum* and *Grastidium* were found to be represented by a single species each. Some species like *D. chrysanthum*, *D. chrysotoxum*, and *D. crepidatum* were very common throughout the state. In contrast, *D. bensoniae, D. capillipes, D. lindleyi, D. salaccense,* and *D. tamenglongense* were rare in the study area in some localized pockets.

Table 4. List of reported *Dendrobium* species not traced in the field condition as well as in the herbaria; only known from earlier literature.

	Scientific name	Reference of the report
1.	*Dendrobium acinaciforme* Roxb.	Deori et al. (2019)
2.	*Dendrobium aduncum* Lindl.	Deori et al. (2019)
3.	*Dendrobium anceps* Sw.	Deori et al. (2019); Kumar & Kumar (2005)
4.	*Dendrobium dantaniense* Guillaumin	Deori et al. (2019)
5.	*Dendrobium farmeri* Paxton	Deori et al.(2019); Kumar & Kumar (2005)
6.	*Dendrobium gratiosissimum* Rchb.f.	Deori et al. (2019); Kumar & Kumar (2005)
7.	*Dendrobium hookerianum* Lindl.	Deori et al. (2019)
8.	*Dendrobium jaintianum* Sabap.	Deori et al. (2019)
9.	*Dendrobium khasianum* Deori	Deori et al. (2019)
10.	*Dendrobium linguella* Rchb.f.	Deori et al. (2019); Kumar & Kumar (2005)
11.	*Dendrobium mannii* Ridl.	Deori et al. (2019)
12.	*Dendrobium numaldeorii* C.Deori, Hynn. & Phukan	Deori et al. (2019)
13.	*Dendrobium peguanum* Lindl.	Deori et al. (2019)
14.	*Dendrobium pendulum* Roxb.	Deori et al. (2019); Kumar & Kumar (2005)
15.	*Dendrobium sulcatum* Lindl.	Deori et al. (2019)
16.	*Dendrobium thyrsiflorum* B.S.Williams	Deori et al. (2019); Kumar & Kumar (2005)

DISCUSSION

Manipur, a part of the Indo-Burma Biodiversity Hotspot (Myers et al. 2000), is one of the orchid rich states in northeastern India. Owing to its geographical location, serene forest cover, and humid climatic conditions, the state is blessed with rich plant genetic resources including orchids. Out of the 67 species reported from the state, only 42 species were seen in the field during this study. Further field surveys are needed to confirm the occurrence of the rest of the 25 species in the state.

Epiphytic *Dendrobium* species are found growing on trunks of small and large trees in tropical and sub-tropical forests in association with other orchid species viz. *Bulbophyllum candidum, B. careyanum, B. cariniflorum, B. gamblei, B. sunipia, Coelogyne corymbosa, C. griffithii, C. nitida, Cymbidium aloifolium, C. erythraeum, Pholidota articulata, P. imbricata, Pinalia acervata*, and *P. amica*.

Few *Dendrobium* species, viz., *Dendrobium aphyllum*, *D. crepidatum, D. devonianum, D. fimbriatum* and *D. nobile* grew as epiphytic as well as lithophytic conditions in tropical and sub-tropical forests.

Image 2. A—*Dendrobium aphyllum* | B—*Dendrobium bensoniae* | C—*Dendrobium cumulatum* | D—*Dendrobium draconis* | E—*Dendrobium lindleyi* | F—*Dendrobium longicornu* | G—*Dendrobium parcum* | H—*Dendrobium parishii* | I—*Dendrobium salaccense* | J—*Dendrobium sinominutiflorum* | K—*Dendrobium stuposum* | L—*Dendrobium tamenglongense*. © H. Bishwajit Sharma.

Some of the host tree species of *Dendrobium*s in Manipur are *Artocarpus chaplasha*, *Bauhinia purpurea*, *Mangifera indica*, *Michelia champaca*, *Quercus serreta*, *Schima wallichii, Terminalia elliptica*, and *Toona ciliata*.

Loss of natural habitats particularly due to tree felling, shifting (Jhum) cultivation practices, construction of hydro-electric dam, railway tracks, and other urbanization practices cause rapid loss of plant genetic resources. As most of the species are epiphytic, illegal trade of timber species also affect their natural habitats resulting in their extermination from the field. So, for survival of the species, continuous monitoring is mandatory at regular interval. As most of the species are very showy, ex situ cultivation is suggested for revenue generation.

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Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23575–23586
ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)
https://doi.org/10.11609/jott.8469.15.7.23575-23586
#8469 | Received 04 April 2023 | Final received 22 June 2023 | Finally accepted 02 July 2023

OPEN ACCESS

COMMUNICATION

Status of macrofungal diversity in the wet evergreen forests of Agasthyamala Biosphere Reserve, Western Ghats, India

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Abstract: Agasthyamala Biosphere Reserve is a part of Western Ghats (India), has diverse ecosystems and constitutes an important biogeographical 'hotspot' which is well known for its species richness and endemism. Since limited information was available on the mycoflora in this area, a survey was conducted to evaluate the macrofungal diversity in the wet evergreen forests of the Agasthyamala Biosphere Reserve. The survey was carried out during the monsoon and post-monsoon seasons of 2021–2022 and revealed the existence of 62 macrofungal species belonging to 43 genera, 24 families, and eight orders. Out of the eight orders, seven orders belong to the division Basidiomycota and the other order Xylariales belongs to Ascomycota. The family Polyporaceae was identified as the dominant family. The survey also noted the presence of saprotrophic and mycorrhizal fungi. Among the identified species, the maximum density was of *Panellus pusillus* (6.08) followed by *Microporus xanthopus* (5.38). *Microporus xanthopus* (82.14%) exhibited the maximum frequency of occurrence and was identified as the most common species. *Coprinellus disseminatus* was the most abundant species among macrofungi. The assessment of macrofungal diversity using the Shannon biodiversity index resulted in a value of 2.99, indicating a rich and diverse fungal population within the forest. This finding emphasizes the significant role of the forest ecosystem in supporting a wide variety of fungi

Keywords: *Ascomycetes*, *basidiomycetes*, endemism, hotspot, Kerala, mushroom diversity, mycoflora.

Editor: Anonymity requested. **Date of publication:** 26 July 2023 (online & print)

Citation: Akshaya, K.K., A. Karthikeyan & C. Kunhikannan (2023). Status of macrofungal diversity in the wet evergreen forests of Agasthyamala Biosphere Reserve, Western Ghats, India. *Journal of Threatened Taxa* 15(7): 23575–23586. https://doi.org/10.11609/jott.8469.15.7.23575-23586

Funding: Indian Council of Forestry Research and Education (ICFRE), Dehra Dun & Compensatory afforestation fund management and planning authority (Project No. AICRP 31), MoEF & CC, Govt of India.

Competing interests: The authors declare no competing interests.

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Author contributions: KKA has done the survey of macrofungi in Agasthayamala forests of Western Ghats of India and also identified the macrofungi. She has written the full part of this manuscript with able guidance of AK. CK helped to identify the plants of Agasthyamala forests and reviewed this manuscript prior to submission.

Acknowledgements: The authors thank Indian Council of Forestry Research and Education, Dehra Dun, India for providing necessary facilities to undertake this study under the funding support of 'Strengthening forestry research for ecological sustainability and productivity enhancement' scheme by Compensatory Afforestation Fund Management and Planning Authority, Ministry of Environment, Forest & Climate change, Government of India vide Project No. AICRP-31. Authors also thank the Department of Forests & Wildlife, Government of Kerala, India for the necessary permission to take up this study.

INTRODUCTION

Fungi represent a distinct and diverse group of organisms that play a crucial role in ecosystem functioning by participating in various ecological cycles (Schmit & Mueller 2007). They constitute one of the largest communities after insects, highlighting their ecological significance. Fungi exhibit remarkable adaptability in terms of morphology, lifestages, developmental patterns, and habitats. They are capable of colonizing a wide range of environments, including those characterized by extreme conditions such as low or high temperatures, and high concentrations of metals and salts (Cox 2007).

Macrofungi represent a prominent group found in forest ecosystems, within the fungal kingdom. The fruiting bodies of macrofungi, belonging to the phyla Ascomycota and Basidiomycota, can be epigeous (aboveground) or hypogeous (underground) (Chang & Miles 1992). These organisms are characterized by their distinct fruiting body forms, which include cup fungi, jelly fungi, coral fungi, polypores, puffballs, corticoid fungi, and agarics.

It has been estimated that the total existence of fungi is about 1.5–5.1 million species, of which approximately 150,000 fungal species have been reported (Blackwell 2011; Berbee et al. 2017; Wu et al. 2019). Macrofungi have a worldwide distribution, ranging 53,000–110,000 species depending on the plant-to-macrofungi ratio (Mueller et al. 2007). In India, the Himalayan and Western Ghats ranges are the major hotspots of fungal diversity (Manoharachary *et al.* 2005).

Western Ghats is one of the biodiversity hotspots of India that covers an area of 160,000 km^2 which extends 1,600 km running parallel to the western coast of the Indian peninsula distributed in six states such as Gujarat, Maharashtra, Goa, Karnataka, Kerala, and Tamil Nadu (UNESCO 2023). Agasthyamala Biosphere Reserve is a part of the Western Ghats, has diverse ecosystems, and constitutes an important biogeographical 'hotspeck'. It is well known for its species richness and endemism. The biosphere reserve is marked by the presence of dominant vegetation like *Palaquium ellipticum* (Dalz.) Baill., *Cullenia exarillata* Robyns, *Elaeocarpus munroii* (Wight) Mast., *Elaeocarpus tuberculatus* Roxb., *Gluta travancorica* Bedd., *Syzygium mundagam* (Bourd.), *Baccaurea courtallensis* (Wight) Mull.Arg., and *Ixora agasthyamalayana* Sivad. & N.Mohanan. The evergreen forests of the biosphere reserve are endowed with many endemics such as *Garcinia travancorica* Bedd., *Garcinia imberti* Bourd., *Humboldtia unijuga* var. *unijuga* Bedd. & var. *trijuga* Joseph & Chandr., and *Syzygium bourdilloni* (Gamble) Rathakr. & Nair (Mohanan & Sivadasan 2002).

Several studies have been carried out in Western Ghats focusing on the diversity, distribution, taxonomy, ecology, nutritional, and bioactive potential of macrofungi (Manimohan & Leelavathy 1988, 1989a,b; Manimohan et al. 1995, 2004, 2007; Pradeep & Vrinda 2010; Puthusseri et al. 2010; Sudheep & Sridhar 2014; Pavithra et al. 2016). The diversity and distribution of macrofungi were investigated by several researchers (Natarajan 1995; Manimohan et al. 2007; Pradeep et al. 2013, 2016). Studies on ectomycorrhizal fungi were conducted by Natarajan & Raman (1983), Mohan (2008), and Mohanan (2014). Hosagoudar & Thomas (2010) conducted a study on foliicolous fungal flora in the Peppara and Neyyar Wildlife Sanctuaries of Agasthyamala Biosphere Reserve. However, the status of macrofungal diversity in the wet evergreen forests of Agasthyamala is limited and requires further investigation and documentation. The present study aims to assess the status of macrofungal diversity in the Agasthyamala Biosphere Reserve area.

MATERIALS & METHODS

Study area

The Agasthyamala Biosphere Reserve is situated at the southernmost tip of the Western Ghats mountain range. Its geographic coordinates range from 8.1333°–9.1666° N & 76.8666°–77.5666° E. Established in 2001, the biosphere reserve covers a total area of 3,500 km^2, with 1,828 km^2 falling within the state of Kerala and 1,672 km^2 within Tamil Nadu. The Agasthyamala Biosphere Reserve encompasses various districts, including Kollam and Thiruvananthapuram in Kerala, and Tirunelveli and Kanyakumari in Tamil Nadu. Within the Kerala region of the reserve, it comprises the Neyyar, Peppara, and Shendurney wildlife sanctuaries, as well as areas such as Achankovil, Thenmala, Konni, Punalur, Thiruvananthapuram Division, and Agasthyavanam Special Division. The region experiences temperatures ranging 18–35°C, and an annual rainfall of 2,400–3,500 mm (Manju et al. 2009).

Survey of macrofungi

The survey was conducted during the monsoon and post-monsoon seasons of 2021–2022 in the wet evergreen forest areas of the Agasthyamala Biosphere Reserve, specifically in the Paruthipally range of Thiruvananthapuram Division, Kerala. The macrofungal

Image 1. Location of study sites on Google Earth.

assessment was carried out using the quadrat method, as described by Harsh (2021). A total of 28 quadrats, each measuring 10 × 10 m, were established in various locations within the forest area. These locations included 36 Mala, Bonacaud Division, Bonacaud, Bonacaud camp shed, Bonacaud School, Cardamom Estate, Elakkad 50 ha, GB Division, Bonacaud Ghost Bungalow, Kallar, Kilavanthottam, Kurushumala, Kurushumala Gate, Pandimotta, Pandipathu, and Bonacaud Picket Station. The selected quadrats exhibited an altitudinal range of 343–1,032 m, as indicated in Table 1 and Image 1.

Macrofungal collection and identification

Macrofungi were photographically recorded in their habitats and the fresh samples were collected with great care in a thermocol box. The macroscopic and ecological characteristics were documented during the collection. A spore print was taken for fleshy mushrooms and noted its color. The samples were dried in a hot air oven at 50 ^{o}C for seven hours and were stored in dry paper covers and labeled with collection numbers for future reference. Specimens were identified with the help of manuals, available literature (Christensen 1968; Ryvarden & Johansen 1980) and online resources like mushroomexpert.com. The nomenclature of the species name is in accordance with the MycoBank database (accessed on 20 January 2023).

Data analysis

The number of sporocarps of each macrofungus in the 10 × 10 m quadrat was enumerated. The quantitative analysis such as frequency, density, and abundance of macrofungal species was calculated by Mishra (1968) as follows:

a. Density: It refers to the numerical strength of a species, and can be calculated using the formula:

$$\text{Density} = \frac{\text{Total number of individuals of a fungal species in all quadrats}}{\text{The total area of the quadrat studied}}$$

b. Frequency (%): Frequency is the degree of dispersion of individual species in an area, which is calculated by the equation:

$$\text{Frequency (\%)} = \frac{\text{Number of quadrats in which the species occurred x 100}}{\text{Total number of quadrats studied}}$$

c. Abundance: indicates the number of individuals

Table 1. Geographic co-ordinates of study sites in Agasthyamala forests.

	Locations	Latitude	Longitude	Altitude
1	36 Mala 10 acre	N 8.69218333°	E 77.18296944°	736 m
2	36 Mala	N 8.67951667°	E 77.18054167°	999 m
3	BA Division	N 8.69185556°	E 77.16231944°	770 m
4	Bonacaud 1	N 8.67007778°	E 77.15408333°	406 m
5	Bonacaud 2	N 8.67104444°	E 77.15293889°	343 m
6	Bonacaud Camp Shed 1	N 8.69279167°	E 77.17595278°	805 m
7	Bonacaud Camp Shed 2	N 8.69504167°	E 77.17340000°	740 m
8	Bonacaud Camp shed 3	N 8.69282222°	E 77.17234444°	730 m
9	Bonacaud School	N 8.68537778°	E 77.16697500°	559 m
10	Cardamom Estate	N 8.68565000°	E 77.18243333°	895 m
11	Elakkad 50 ha	N 8.68229444°	E 77.17858333°	695 m
12	GB Division	N8.69356667°	E 77.17162778°	635 m
13	Ghost House	N 8.69027222°	E 77.16896667°	770 m
14	Kallar 1	N 8.69493333°	E 77.15632222°	648 m
15	Kallar 2	N 8.69384167°	E 77.15466944°	635 m
16	Kallar 3	N 8.69280000°	E 77.15594444°	623 m
17	Kilavanthottam 1	N 8.69196944°	E 77.17507778°	862 m
18	Kilavanthottam 2	N8.69220556°	E 77.17670556°	882 m
19	Kilavanthottam 3	N 8.69315000°	E 77.17493056°	829 m
20	Kilavanthottam 4	N 8.69198889°	E 77.17819167°	947 m
21	Kurushumala	N 8.68520833°	E 77.15527222°	719 m
22	Kurushumala Gate	N 8.68521389°	E 77.15784167°	690 m
23	Pandimotta	N 8.69136389°	E 77.18407222°	1,004 m
24	Pandipath 1	N 8.68534167°	E 77.18038889°	1,032 m
25	Pandipath 2	N 8.68967222°	E 77.18673056°	989 m
26	Picket Station 1	N 8.66555000°	E 77.17373611°	598 m
27	Picket Station 2	N 8.66408056°	E 77.17267778°	546 m
28	Picket station 3	N 8.66590000°	E 77.17113333°	574 m

of different species in the community per unit area. It is calculated by the equation:

$$\text{Abundance} = \frac{\text{Total number of individuals of a species in all quadrats}}{\text{Total number of quadrats in which the species present}}$$

Species diversity analysis

The macrofungal diversity in different forest areas of Agasthyamala Biosphere Reserve was determined using the Shannon diversity index Magurran (1988).

Shannon diversity index, $H' = -\sum p_i \ln p_i$

Where pi is the proportion of individuals of a species to the total number of species.

RESULTS

Macrofungal assessment

A comprehensive assessment of macrofungal diversity in the present study revealed the presence of 62 macrofungal species, representing 43 genera, 24 families, and eight orders. Among these, seven orders belonged to the division Basidiomycota, while one order belonged to Ascomycota. The dominant order observed was Agaricales, comprising 33 species, followed by Polyporales with 13 species. Auriculariales accounted for seven species, while Dacrymycetales, Russulales, Tremellales, and Xylariales each had two species, and Boletales had single species (Table 2, Figure 1). The family Polyporaceae exhibited the highest species richness, with nine species recorded, while families such as Schizophyllaceae, Mycenaceae, Serpulaceae, Russulaceae, and Hypoxylaceae displayed a lower number of macrofungi (Table 2).

Ecological preference of macrofungi

The present study revealed that the maximum numbers of species (55) were saprophytes and a few species were mycorrhizal (seven) in nature. Mycorrhizal macrofungi were found in the soil, associated with the roots of higher angiosperm species (Table 3).

Quantitative analysis

The quantitative study of the macrofungal species showed that maximum density was represented by the species *Panellus pusillus* (6.08), followed by *Microporus xanthopus* (5.38). The maximum frequency of occurrence was exhibited by *Microporus xanthopus* (82.14%), followed by *Stereum ostrea* (42.86%), *Auricularia delicata* (32.14%), and *Dacryopinax spathularia* (25%). *Microporus xanthopus* was the most frequent species present in the Agasthyamala forests. *Coprinellus disseminatus* was the most abundant species of macrofungi (Table 4).

Species diversity analysis

The species diversity of macrofungi was calculated using the Shannon diversity index. The Shannon diversity index for macrofungi in the wet evergreen forests of Agasthyamala was found to be 2.99 (Table 5).

The current study enumerated a total of 62 macrofungal species which are edible, medicinal,

Table 2. Distribution of macrofungi in their respective family and order.

	Species	Family	Order
1	*Amanita vaginata* (Bull.) Lam.	Amanitaceae	Agaricales
2	*Amauroderma rugosum* (Blume & T.Nees) Torrend	Ganodermataceae	Poyporales
3	*Anthracophyllum archeri* (Berk.) Pegler	Omphalotaceae	Agaricales
4	*Anthracophyllum* sp.	Omphalotaceae	Agaricales
5	*Auricularia delicata* (Fr.) heim	Auriculariaceae	Auriculariales
6	*Auricularia mesenterica* (Dicks.) Pers.	Auriculariaceae	Auriculariales
7	*Auricularia* sp. 1	Auriculariaceae	Auriculariales
8	*Auricularia* sp. 2	Auriculariaceae	Auriculariales
9	*Auricularia* sp. 3	Auriculariaceae	Auriculariales
10	*Campanella caesia* Romagn.	Marasmiaceae	Agaricales
11	*Campanella tristis* (G. Stev.) Segedin	Marasmiaceae	Agaricales
12	*Chlorophyllum molybdites* (G. Mey.) Massee	Agaricaceae	Agaricales
13	*Clavulinopsis fusiformis* (Soweby) Corner	Clavariaceae	Agaricales
14	*Coprinellus domesticus* (Bolton) Vilgalys, Hopple & Jacq.Johnson	Psathyrellaceae	Agaricales
15	*Coprinellus disseminatus* (Pers.) J.E.Lange	Psathyrellaceae	Agaricales
16	*Crepidotus variabilis* (Pers.) P. Kumm.	Crepidotaceae	Agaricales
17	*Cyptotrama asprata* (Berk.) Redhead & Ginns	Physalacriaceae	Agaricales
18	*Cuphophyllus pratensis* (Schaeff.) Bon	Hygrophoraceae	Agaricales
19	*Dacrymyces palmatus* (Schwein.) Burt	Dacryomycetaceae	Dacrymycetales
20	*Dacryopinax spathularia* (Schwein) G.W.Martin	Dacryomycetaceae	Dacrymycetales
21	*Daedaleopsis confragosa* (Bolton) J.Schrot	Fomitopsidaceae	Polyporales
22	*Daldinia concentrica* (Bolton) Ces. & De.Not.	Hypoxylaceae	Xylariales
23	*Earliella scabrosa* (Pers.) Glib. & Ryvarden	Polyporaceaea	Polyporales
24	*Exidia glandulosa* (Bull.) Fr.	Auriculariaceae	Auriculariales
25	*Exidia recisa* Ditmar (Fr.)	Auriculariaceae	Auriculariales
26	*Ganoderma applanatum* (Pers.) Pat.	Ganodermataceae	Polyporales
27	*Ganoderma* sp.	Ganodermataceae	Polyporales
28	Gill fungi 1	Marasmiaceae	Agaricales
29	Gill fungi 2	Marasmiaceae	Agaricales
30	*Hexagonia tenuis* (Hook.) Fr.	Polyporaceaea	Polyporales
31	*Hymenopellis radicata* (Relhan) R.H.Petersen	Physalacriaceae	Agaricales
32	*Lentinus* sp. 1	Agaricaceae	Agaricales
33	*Lentinus* sp. 2	Agaricaceae	Agaricales
34	*Lentinus* sp. 3	Agaricaceae	Agaricales
35	*Lentinus tigrinus* (Bull.) Fr.	Agaricaceae	Agaricales
36	*Lenzites acuta* Berk.	Polyporaceaea	Polyporales
37	*Leucoagaricus rubrotinctus* (Peck) Singer	Agaricaceae	Agaricales
38	*Leucocoprinus fragilissimus* (Berk. & M.A.Curtis) Pat.	Polyporaceaea	Polyporales
39	*Macrolepiota procera* (Scop.) Singer	Agaricaceae	Agaricales
40	*Marasmiellus* sp.	Marasmiaceae	Agaricales
41	*Marasmius haematocephalus* (Mont.) Fr.	Marasmiaceae	Agaricales
42	*Marasmius siccus* (Schwein.) Fr.	Marasmiaceae	Agaricales
43	*Marasmius* sp.	Marasmiaceae	Agaricales
44	*Microporellus dealbatus* (Berk. & M.A.Curtis)	Polyporaceaea	Polyporales
45	*Microporus xanthopus* (Fr.) Kuntze.	Polyporaceaea	Polyporales
46	*Mucronella bresadolae*(Quel.) Corner	Clavariaceae	Agaricales
47	*Panellus pusillus* (Pers. Ex Lev.) Burds. & O.K.Mill.	Mycenaceae	Agaricales
48	*Pleurotus* sp. 1	Pleurotaceae	Agaricales
49	*Pleurotus* sp. 2	Pleurotaceae	Agaricales
50	*Pleurotus* sp. 3	Pleurotaceae	Agaricales
51	*Pleurotus* sp. 4	Pleurotaceae	Agaricales
52	*Polyporus grammocephalus* Berk.	Polyporaceaea	Polyporales
53	*Pycnoporus sanguineus* (L.) Murrill	Polyporaceaea	Polyporales
54	*Russula cyanoxantha* (Schaeff.) Fr.	Russulaceae	Russulales
55	*Schizophyllum commune* Fr.	Schizophyllaceae	Agaricales
56	*Serpula similis* (Berk. & Broome) Ginns	Serpulaceae	Boletales
57	*Stereum ostrea* (Blume & T.Nees) Fr.	Stereaceae	Russulales
58	*Termitomyces microcarpus* (Berk. & Broome) R.Heim	Lyophyllaceae	Agaricales
59	*Trametes gibbosa* (Pers.) Fr.	Polyporaceaea	Polyporales
60	*Tremella fuciformis* Berk.	Tremellaceae	Tremellales
61	*Tremella mesenterica* Retz.	Tremellaceae	Tremellales
62	*Xylaria longipes* Nitschke	Xylariaceae	Xylariales

Table 3. List of macrofungi recorded in the forests of Agasthyamala with their habitat, mode of nutrition and associate.

	Species	Habitat	Nutrition	*Associate
1	*Amanita vaginata* (Bull.) Lam.	Soil	Mycorrhizal	+
2	*Amauroderma rugosum* (Blume & T.Nees) Torrend	Soil	Saprotrophic	-
3	*Anthracophyllum archeri* (Berk.) Pegler	Dead twig	Saprotrophic	-
4	*Anthracophyllum* sp.	Dead twig	Saprotrophic	-
5	*Auricularia delicata* (Fr.) Heim	Dead wood	Saprotrophic	-
6	*Auricularia mesenterica* (Dicks.) Pers.	*Gordonia obtusa*	Saprotrophic	+
7	*Auricularia* sp. 1	Deadwood	Saprotrophic	-
8	*Auricularia* sp. 2	Deadwood	Saprotrophic	-
9	*Auricularia* sp. 3	Deadwood	Saprotrophic	-
10	*Campanella caesia* Romagn.	Fallen twig	Saprotrophic	-
11	*Campanella tristis* (G.Stev.) Segedin	Deadwood	Saprotrophic	-
12	*Chlorophyllum molybdites* (G.Mey.) Massee	Soil	Saprotrophic	-
13	*Clavulinopsis fusiformis* (Soweby) Corner	Soil	Saprotrophic	-
14	*Coprinellus domesticus* (Bolton) Vilgalys, Hopple & Jacq.Johnson	Deadwood	Saprotrophic	-
15	*Coprinellus disseminatus* (Pers.) J.E.Lange	Soil	Saprotrophic	-
16	*Crepidotus variabilis* (Pers.) P.Kumm.	*Gordonia obtusa*	Saprotrophic	+
17	*Cyptotrama asprata* (Berk.) Redhead & Ginns	Soil	Saprotrophic	-
18	*Cuphophyllus pratensis* (Schaeff.) Bon	Soil	Mycorrhizal	+
19	*Dacrymyces palmatus* (Schwein.) Burt	Dead wood	Saprotrophic	-
20	*Dacryopinax spathularia* (Schwein) G.W.Martin	Deadwood	Saprotrophic	-
21	*Daedaleopsis confragosa* (Bolton) J.Schrot	Dead wood	Saprotrophic	-
22	*Daldinia concentrica* (Bolton) Ces. & De.Not.	Deadwood	Saprotrophic	-
23	*Earliella scabrosa* (Pers.) Glib. & Ryvarden	Dead wood	Saprotrophic	-
24	*Exidia glandulosa* (Bull.) Fr.	Dead wood	Saprotrophic	-
25	*Exidia recisa* Ditmar (Fr.)	Dead wood	Saprotrophic	-
26	*Ganoderma applanatum* (Pers.) Pat.	Dead wood	Saprotrophic	-
27	*Ganoderma* sp.	Dead wood	Saprotrophic	-
28	Gill fungi 1	Dead wood	Saprotrophic	-
29	Gill fungi 2	Dead wood	Saprotrophic	-
30	*Hexagonia tenuis* (Hook.) Fr.	Dead wood	Saprotrophic	-
31	*Hymenopellis radicata* (Relhan) R.H.Petersen	Soil	Saprotrophic	-
32	*Lentinus* sp. 1	Dead wood	Saprotrophic	-
33	*Lentinus* sp. 2	Dead wood	Saprotrophic	-
34	*Lentinus* sp. 3	Dead wood	Saprotrophic	-
35	*Lentinus tigrinus* (Bull.) Fr.	Dead wood	Saprotrophic	-
36	*Lenzites acuta* Berk.	Dead wood	Saprotrophic	-
37	*Leucoagaricus rubrotinctus* (Peck) Singer	Soil	Mycorrhizal	+
38	*Leucocoprinus fragilissimus* (Berk. & M.A.Curtis) Pat.	Soil	Saprotrophic	
39	*Macrolepiota procera* (Scop.) Singer	Soil	Mycorrhizal	+
40	*Marasmiellus* sp.	Dead wood	Saprotrophic	-
41	*Marasmius haematocephalus* (Mont.) Fr.	Dead wood	Saprotrophic	-
42	*Marasmius siccus* (Schwein.) Fr.	Dead wood	Saprotrophic	-
43	*Marasmius* spp.	Dead wood	Saprotrophic	-
44	*Microporellus dealbatus* (Berk. & M.A.Curtis)	Dead wood	Saprotrophic	-
45	*Microporus xanthopus* (Fr.) Kuntze.	Dead fallen twig	Saprotrophic	-
46	*Mucronella bresadolae* (Quel.) Corner	*Ficus exasperata*	Saprotrophic	+
47	*Panellus pusillus* (Pers. Ex Lev.) Burds. & O.K.Mill.	Dead wood	Saprotrophic	-
48	*Pleurotus* sp. 1	Dead wood	Saprotrophic	-
49	*Pleurotus* sp. 2	Dead wood	Saprotrophic	-
50	*Pleurotus* sp. 3	Dead wood	Saprotrophic	-
51	*Pleurotus* sp. 4	Dead wood	Saprotrophic	-
52	*Polyporus grammocephalus* Berk.	Dead wood	Saprotrophic	-
53	*Pycnoporus sanguineus* (L.) Murrill	Dead wood	Saprotrophic	-
54	*Russula cyanoxantha* (Schaeff.) Fr.	Soil	Mycorrhizal	+
55	*Schizophyllum commune* Fr.	Dead wood	Saprotrophic	-
56	*Serpula similis* (Berk. & Broome) Ginns	*Elaeocarpus munroii*	Mycorrhizal	+
57	*Stereum ostrea* (Blume & T.Nees) Fr.	Dead wood	Saprotrophic	-
58	*Termitomyces microcarpus* (Berk. & Broome) R.Heim	Soil	Mycorrhizal	-
59	*Trametes gibbosa* (Pers.) Fr.	Dead wood	Saprotrophic	-
60	*Tremella fuciformis* Berk.	Fallen twig	Saprotrophic	-
61	*Tremella mesenterica* Retz.	Dead wood	Saprotrophic	-
62	*Xylaria longipes* Nitschke	Dead wood	Saprotrophic	-

*: (+)—associated with tree| (-)—non associated with tree.

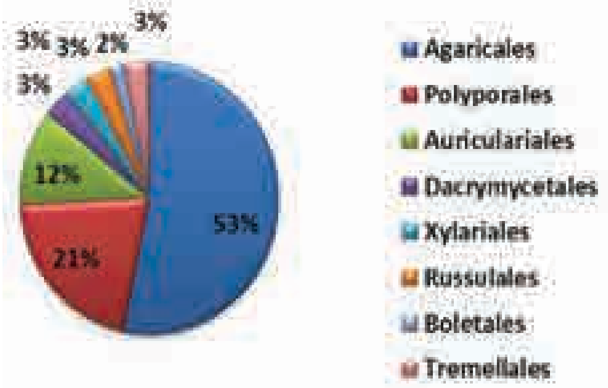

Figure 1. Percentage of distribution of macrofungi in different orders.

ectomycorrhizal, saprotrophic and toxic (e.g., *Chlorophyllum molybdites*) belonging to 43 genera, 24 families and eight orders from Agasthyamala Forests. These forests provide fairly undisturbed natural habitats for a variety of macrofungi. Among them, seven orders belong to Basidiomycota and one order, Xylariales, comes under Ascomycota. Polyporaceae is the largest family with nine genera followed by Marasmiaceae (Eight genera), Agaricaceae and Auriculariaceae (Seven genera each), Pleurotaceae (Four genera), Ganodermataceae (Three genera), Omphalotaceae, Clavariaceae, Physalacriaceae, Dacryomycetaceae and Tremellaceae (Two genera each) and all remaining families like Schizophyllaceae, Mycenaceae, Coprinaceae, Serpulaceae were represented by a single genus each. The order Agaricales was dominant (33 species), followed by Polyporales (13 species), Auriculariales (Seven species), Dacrymycetales, Russulales, Tremellales, and Xylariales each having two species and Boletales having only one species.

The quantitative analysis of macrofungi revealed that *Panellus pusillus* (6.08) showed the maximum density followed by *Microporus xanthopus* (5.38). The maximum frequency of occurrence was exhibited by *Microporus xanthopus* (82.14%) followed by *Stereum ostrea* (42.86%), *Auricularia delicata* (32.14%) and *Dacryopinax spathularia* (25%). *Microporus xanthopus was identified as the most common species in the forests of* Agasthyamala. *Coprinellus disseminatus* was the most abundant species. The Shannon diversity index for macrofungi was calculated and found to be 2.99, indicating rich fungal biodiversity and less human interference in this area.

DISCUSSION

The Indian region is renowned for hosting four biodiversity hotspots. The Western Ghats and Sri Lanka are renowned biodiversity hotspots, attracting scientific exploration due to their rich variety of life forms. Among these, macrofungi, including mushrooms and polypores, hold immense importance as edible and medicinal species, with significant bio-prospecting potential. Additionally, macrofungi play a crucial role in ecosystem functioning by aiding in the formation of humus and nutrient recycling on the forest floor. Although previous studies have reported a wide range of agarics and other fungi from the evergreen and semi-evergreen forests of the Nilgiri Biosphere Reserve and the Kodagu region, further research is necessary to comprehensively document the macrofungal diversity in these areas.

Several studies have been conducted in the region, including those by Tapwal et al. (2013), Gogoi & Prakash (2015), and Vishwakarma et al. (2017), which are similar in nature. The findings of our study align with previous research, particularly regarding the dominance of the Agaricales order, as reported by Tapwal et al. (2013). Senthilarasu (2014) observed that Agaricales is the most dominant order, followed by Polyporales. Gogoi & Prakash (2015) reported the highest number of Agaricales. Our study confirms the ecological preference of the species, with the majority being saprophytic and a few exhibiting mycorrhizal characteristics, consistent with the findings of Tapwal et al. (2013). Notable macrofungi such as *Amanita vaginata*, *Cuphophyllus pratensis*, *Leucoagaricus rubrotinctus*, *Macrolepiota procera*, *Russula cyanoxantha*, *Serpula similis*, and *Termitomyces microcarpus* were identified as ectomycorrhizal fungi, known to enhance soil nutrient status, water availability, and disease resistance (Waring & Running 2007; Harsh 2021). The symbiotic association between fungi and plant roots contributes to the survival, growth, and development of plants, aiding in the absorption of minerals, particularly phosphate and water, from the soil (Jorgensen & Shoulders 1967; Marks & Kozlowski 1973; Onguene & Kuyper 2001). The extensive underground network of mycorrhizal fungal hyphae enhances the overall well-being of the ecosystem, making mycorrhiza the most efficient nutrient uptake system in nature (Onguene & Kuyper 2001). The presence of both saprophytic and mycorrhizal fungi in the study area indicates a healthy condition of the forest (Tapwal et al. 2013).

Macrofungi that inhabit woody substrates can be categorized as either saprophytic or plant pathogenic,

Table 4. Density, frequency and abundance of macrofungi of Agasthyamala forests.

	Species	Density	Frequency (%)	Abundance
1	*Amanita vaginata* (Bull.) Lam.	0.03	7.14	1.50
2	*Amauroderma rugosum* (Blume & T.Nees) Torrend	0.01	3.57	1.00
3	*Anthracophyllum archeri* (Berk.) Pegler	2.69	17.86	53.80
4	*Anthracophyllum* sp.	2.21	14.29	55.25
5	*Auricularia delicata* (Fr.) Heim	2.39	32.14	26.56
6	*Auricularia mesenterica* (Dicks.) Pers.	0.24	7.14	12.00
7	*Auricularia* sp. 1	0.4	3.57	40.00
8	*Auricularia* sp. 2	0.15	3.57	15.00
9	*Auricularia* sp. 3	0.35	3.57	35.00
10	*Campanella caesia* Romagn.	0.03	3.57	3.00
11	*Campanella tristis* (G.Stev.) Segedin	0.4	14.29	10.00
12	*Chlorophyllum molybdites* (G.Mey.) Massee	0.03	3.57	3.00
13	*Clavulinopsis fusiformis* (Soweby) Corner	0.06	3.57	6.00
14	*Coprinellus domesticus* (Bolton) Vilgalys, Hopple & Jacq.Johnson	1.34	10.71	44.67
15	*Coprinellus disseminatus* (Pers.) J.E.Lange	0.6	3.57	60.00
16	*Crepidotus variabilis* (Pers.) P.Kumm.	0.14	10.71	4.67
17	*Cyptotrama asprata* (Berk.) Redhead & Ginns	0.05	14.29	1.25
18	*Cuphophyllus pratensis* (Schaeff.) Bon	0.04	7.14	2.00
19	*Dacrymyces palmatus* (Schwein.) Burt	1.19	14.29	29.75
20	*Dacryopinax spathularia* (Schwein) G.W.Martin	2.42	25.00	34.57
21	*Daedaleopsis confragosa* (Bolton) J.Schrot	0.13	14.29	3.25
22	*Daldinia concentrica* (Bolton) Ces. & De.Not.	0.02	7.14	1.00
23	*Earliella scabrosa* (Pers.) Glib. & Ryvarden	1.02	17.86	20.40
24	*Exidia glandulosa* (Bull.) Fr.	0.01	3.57	1.00
25	*Exidia recisa* Ditmar (Fr.)	0.11	7.14	5.50
26	*Ganoderma applanatum* (Pers.) Pat.	0.05	7.14	2.50
27	*Ganoderma* sp.	0.07	7.14	3.50
28	Gill fungi 1	0.35	3.57	35.00
29	Gill fungi 2	0.15	3.57	15.00
30	*Hexagonia tenuis* (Hook.) Fr.	0.6	32.14	6.67
31	*Hymenopellis radicata* (Relhan) R.H.Petersen	0.01	3.57	1.00
32	*Lentinus* sp. 1	0.21	3.57	21.00
33	*Lentinus* sp. 2	0.41	3.57	41.00
34	*Lentinus* sp. 3	0.02	3.57	2.00
35	*Lentinus tigrinus* (Bull.) Fr.	0.04	3.57	4.00
36	*Lenzites acuta* Berk.	0.23	17.86	4.60
37	*Leucoagaricus rubrotinctus* (Peck) Singer	0.05	7.14	2.50
38	*Leucocoprinus fragilissimus* (Berk. & M.A.Curtis) Pat.	0.01	3.57	1.00
39	*Macrolepiota procera* (Scop.) Singer	0.01	3.57	1.00
40	*Marasmiellus* sp.	0.25	7.14	12.50
41	*Marasmius haematocephalus* (Mont.) Fr	0.1	17.86	2.00
42	*Marasmius siccus* (Schwein.) Fr.	0.03	3.57	3.00
43	*Marasmius* spp.	0.04	7.14	2.00
44	*Microporellus dealbatus* (Berk. & M.A.Curtis)	0.01	3.57	1.00
45	*Microporus xanthopus* (Fr.) Kuntze.	5.38	82.14	23.39
46	*Mucronella bresadolae*(Quel.) Corner	0.27	3.57	27.00
47	*Panellus pusillus* (Pers. Ex Lev.) Burds. & O.K.Mill.	6.08	28.57	76.00
48	*Pleurotus* sp. 1	0.07	3.57	7.00
49	*Pleurotus* sp. 2	0.07	3.57	7.00
50	*Pleurotus* sp. 3	0.01	3.57	1.00
51	*Pleurotus* sp. 4	0.02	3.57	2.00
52	*Polyporus grammocephalus* Berk.	0.01	3.57	1.00
53	*Pycnoporus sanguineus* (L.) Murrill	0.79	10.71	26.33
54	*Russula cyanoxantha* (Schaeff.) Fr.	0.01	3.57	1.00
55	*Schizophyllum commune* Fr.	0.89	10.71	29.67
56	*Serpula similis* (Berk. & Broome) Ginns	0.09	3.57	9.00
57	*Stereum ostrea* (Blume & T.Nees) Fr.	1.34	42.86	11.17
58	*Termitomyces microcarpus* (Berk. & Broome) R.Heim	1.79	14.29	44.75
59	*Trametes gibbosa* (Pers.) Fr.	0.02	7.14	1.00
60	*Tremella fuciformis* Berk.	0.01	3.57	1.00
61	*Tremella mesenterica* Retz.	0.33	7.14	16.50
62	*Xylaria longipes* Nitschke	0.01	3.57	1.00

Table 5. Diversity of macrofungal species by Shannon index.

	Species	Density	Pi ln pi
1	*Amanita vaginata* (Bull.) Lam.	0.03	-0.01
2	*Amauroderma rugosum* (Blume & T.Nees) Torrend	0.01	0.00
3	*Anthracophyllum archeri* (Berk.) Pegler	2.69	-0.19
4	*Anthracophyllum* spp.	2.21	-0.17
5	*Auricularia delicata* (Fr.) Heim	2.39	-0.18
6	*Auricularia mesenterica* (Dicks.) Pers.	0.24	-0.03
7	*Auricularia* sp. 1	0.4	-0.05
8	*Auricularia* sp. 2	0.15	-0.02
9	*Auricularia* sp. 3	0.35	-0.05
10	*Campanella caesia* Romagn.	0.03	-0.01
11	*Campanella tristis* (G.Stev.) Segedin	0.4	-0.05
12	*Chlorophyllum molybdites* (G.Mey.) Massee	0.03	-0.01
13	*Clavulinopsis fusiformis* (Soweby) Corner	0.06	-0.01
14	*Coprinellus domesticus* (Bolton) Vilgalys, Hopple & Jacq.Johnson	1.34	-0.12
15	*Coprinellus disseminatus* (Pers.) J.E.Lange	0.6	-0.07
16	*Crepidotus variabilis* (Pers.) P.Kumm.	0.14	-0.02
17	*Cyptotrama asprata* (Berk.) Redhead & Ginns	0.05	-0.01
18	*Cuphophyllus pratensis* (Schaeff.) Bon	0.04	-0.01
19	*Dacrymyces palmatus* (Schwein.) Burt	1.19	-0.11
20	*Dacryopinax spathularia* (Schwein) G.W.Martin	2.42	-0.18
21	*Daedaleopsis confragosa* (Bolton) J.Schrot	0.13	-0.02
22	*Daldinia concentrica* (Bolton) Ces. & De.Not.	0.02	0.00
23	*Earliella scabrosa* (Pers.) Glib. & Ryvarden	1.02	-0.10
24	*Exidia glandulosa*(Bull.) Fr.	0.01	0.00
25	*Exidia recisa* Ditmar (Fr.)	0.11	-0.02
26	*Ganoderma applanatum* (Pers.) Pat.	0.05	-0.01
27	*Ganoderma* sp.	0.07	-0.01
28	Gill fungi 1	0.35	-0.05
29	Gill fungi 2	0.15	-0.02
30	*Hexagonia tenuis* (Hook.) Fr.	0.6	-0.07
31	*Hymenopellis radicata* (Relhan) R.H.Petersen	0.01	0.00
32	*Lentinus* sp. 1	0.21	-0.03
33	*Lentinus* sp. 2	0.41	-0.05
34	*Lentinus* sp. 3	0.02	0.00
35	*Lentinus tigrinus* (Bull.) Fr.	0.04	-0.01
36	*Lenzites acuta* Berk.	0.23	-0.03
37	*Leucoagaricus rubrotinctus* (Peck) Singer	0.05	-0.01
38	*Leucocoprinus fragilissimus* (Berk. & M.A.Curtis) Pat.	0.01	0.00
39	*Macrolepiota procera* (Scop.) Singer	0.01	0.00
40	*Marasmiellus* sp.	0.25	-0.03
41	*Marasmius haematocephalus* (Mont.) Fr	0.1	-0.02
42	*Marasmius siccus* (Schwein.) Fr.	0.03	-0.01
43	spp.	0.04	-0.01
44	*Microporellus dealbatus* (Berk. & M.A.Curtis)	0.01	0.00
45	*Microporus xanthopus* (Fr.) Kuntze.	5.38	-0.28
46	*Mucronella bresadolae*(Quel.) Corner	0.27	-0.04
47	*Panellus pusillus* (Pers. Ex Lev.) Burds. & O.K.Mill.	6.08	-0.30
48	*Pleurotus* sp. 1	0.07	-0.01
49	*Pleurotus* sp. 2	0.07	-0.01
50	*Pleurotus* sp. 3	0.01	0.00
51	*Pleurotus* sp. 4	0.02	0.00
52	*Polyporus grammocephalus* Berk.	0.01	0.00
53	*Pycnoporus sanguineus* (L.) Murrill	0.79	-0.08
54	*Russula cyanoxantha* (Schaeff.) Fr.	0.01	0.00
55	*Schizophyllum commune* Fr.	0.89	-0.09
56	*Serpula similis* (Berk. & Broome) Ginns	0.09	-0.02
57	*Stereum ostrea* (Blume & T.Nees) Fr.	1.34	-0.12
58	*Termitomyces microcarpus* (Berk. & Broome) R.Heim	1.79	-0.15
59	*Trametes gibbosa* (Pers.) Fr.	0.02	0.00
60	*Tremella fuciformis* Berk.	0.01	0.00
61	*Tremella mesenterica* Retz.	0.33	-0.04
62	*Xylaria longipes* Nitschke	0.01	0.00
Shannon diversity index, H' = 2.99			

as documented by Mueller et al. (2007). In the present study, several saprophytic fungi were identified, including *Polyporus grammocephalus*, *Pycnoporus sanguineus*, *Stereum ostrea*, and *Microporellus dealbatus*. Senn-Irlet et al. (2007) reported that approximately 50% of macrofungi found in forests are involved in wood decay processes. Saprophytic macrofungi play a crucial role in carbon and nutrient recycling within ecosystems (Gates, 2009). They can be further classified into three types based on their wood degradation mechanisms: soft rot fungi, white rot fungi, and brown rot fungi. White rot fungi, such as *Pycnoporus sanguineus*, *Trametes gibbosa*, *Earliella scabrosa*, and *Microporellus dealbatus*, are predominant in the studied area. These fungi, belonging to both Ascomycota and Basidiomycota, are responsible for breaking down lignin, cellulose, and hemicellulose in wood. They are unique in their ability to degrade lignin, distinguishing them from other organisms. In contrast, brown rot fungi primarily degrade cellulose and hemicellulose while leaving the lignin relatively intact.

Image 2. a–l Sporocarps of macrofungi within their habitat in the wet evergreen forests of Agasthyamala: a—*Polyporus grammocephalus* | b—*Microporus xanthopus* | c—*Ganoderma applanatum* | d—*Leucoagaricus rubrotinctus* | e—*Hexagonia tenuis* | f—*Dacryopinax spathularia* | g—*Cuphophyllus pratensis* | h—*Mucronella bresadolae* | i—*Pycnoporus sanguineus* | j—*Amanita vaginata* | k—*Cyptotrama asprata* | l—*Macrolepiota procera*. © K.K. Akshaya.

Notably, brown rot fungi like *Dacryopinax spathularia*, were also observed in this study. An interesting characteristic of brown rot fungi is their formation of bracket-shaped fruiting bodies on dead wood.

Termitomyces microcarpus is the most common edible mushroom used by the local people of

Agasthyamala. The local people named the mushroom 'Areekoonu' (Malayalam: rice mushroom). Mushrooms are rich in protein, vitamins, and minerals and are used as a substitute for animal protein (Chang & Buswell 1996).

The sporocarps of macrofungi show diverse forms in their morphology like jelly fungi, polypores, agarics, and coral fungi. The present study reports jelly fungi such as *Tremella fuciformis*, *Exidia recisa*, and *Tremella mesenterica*. Polypores like *Microporus xanthopus,* and *Polyporus grammocephalus,* Agarics such as *Leucoagaricus rubrotinctus*, *Amanita vaginata*, *Chlorophyllum molybdites* and coral fungi include *Clavulinopsis fusiformis*.

The Shannon diversity index was calculated and found to be 2.99, indicating that the study area has high fungal biodiversity. The rich mycoflora of the wet evergreen forests of Agasthyamala is due to less human interference.

The fruiting behavior of macrofungi is influenced by various factors, including elevation, latitude, and their impact on temperature and precipitation (Ohenoja 1993). Macrofungi exhibit distinct patterns of sporocarp production, occurring in different seasons and across extensive geographic areas with notable elevation changes. The presence of specific vegetation types plays a crucial role in determining the species richness and composition of macrofungi in a given area. Grasslands, deserts, forests, tundra, and other habitats harbor characteristic macrofungal species adapted to their respective environments. The abundance and diversity of macrofungi are closely linked to the composition of plant species, as plants serve as vital constituents and energy sources within the ecosystem, supporting the growth and development of most macrofungi. Furthermore, the distribution of ectomycorrhizal fungi, which establish symbiotic relationships with plant roots, often aligns with specific forest types (Natel & Neumann 1992).

The fungal community responds to changes in climatic conditions in the form of changes in fruiting patterns, productivity, fruit body size, geographical distribution, and phenological patterns. Such changes have a strong impact on their functional attributes like modifying carbon cycling, altering bacterial community, and disrupting mycorrhizal associations with effects reflecting up to higher trophic levels. Long-term ecological monitoring studies help to provide valuable insights in ecology, environmental change, natural resource management, and biodiversity conservation. Therefore, understanding the factors that trigger sporocarp community response to climate at species level is very important to predict future species composition and abundance under global climate change scenario.

CONCLUSION

Macrofungi play a crucial role in the ecosystem by significantly influencing soil nutrition, organic carbon levels, and the well-being of surrounding vegetation. Despite the limited availability of reports on macrofungal diversity in the Western Ghats region of India, this study aimed to fill this knowledge gap by generating essential baseline data on higher fungi. The findings of this study revealed that the Agasthyamala Biosphere Reserve exhibits a remarkable richness of fungal diversity, particularly in its wet evergreen forests. The presence of such rich fungal diversity serves as an indicator of the overall health and vitality of these forests. Consequently, the baseline data obtained from this study serves as a valuable resource for understanding and assessing the species richness of macrofungi within these forest ecosystems, contributing to their conservation and management.

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Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23587–23595

ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)

https://doi.org/10.11609/jott.8081.15.7.23587-23595

#8081 | Received 30 June 2022 | Final received 16 May 2023 | Finally accepted 20 June 2023

COMMUNICATION

Developing a fast, reproducible, and simple protocol for virtual lichen herbarium using barcoding and QR code techniques

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Abstract: In recent days, biological specimens are digitalized and digital images are available in virtual herbarium for teaching and learning process. Now, there is a need to explore possibilities of usage of barcodes and quick response (QR) codes in developing virtual herbarium for quick access as well as study the taxonomy of repository specimens. In order to establish a virtual herbarium for lichens using barcode and QR code techniques, lichen specimens such as *Chrysothrix candelaris* (L.) J.R.Laundon, *Leucodermia leucomelos* (L.) Kalb, *Heterodernia flabellate* (fee) D.D.Awasthi, *Parmotrema andinum* (Mull.Arg.) Hale, *Parmotrema grayanum* (Hue) Hale, *Parmellinella stuppeum* (Taylor) Hale, and *Ramalina intermedia* (Delise ex Nyl.) Nyl. were collected from the Eastern Ghats and the Western Ghats of Tamil Nadu, India and were identified based on morphological, anatomical and biochemical methods. Moreover, these specimens were preserved in the conventional lichen herbarium as reference materials for future studies. The barcodes and QR codes were generated for all the repository specimens to access the materials as well as to get a complete description of the lichen specimens. The generated barcodes provided the binomial name of lichen specimens along with their accession number. Similarly, the QR codes provided the digital image of lichen specimens along with complete descriptions such as distribution, habit and habitat, growth forms, name of the family, reproductive structure, chemistry, nature of thallus structure and lichen secondary metabolites. From these studies, we standardized a simple, rapid with reproducible protocol to develop a virtual herbarium for lichens to get the digital image and to access the complete descriptions of lichen specimens. This study might be useful for Lichenologists to get information about lichens in digital form and to maintain the lichen wealth for future regenerations without disturbing the lichen biodiversity.

Keywords: Barcodes, conventional lichen herbarium, digital images, lichen biodiversity, QR codes, quick access, repository, specimens.

Editor: Anonymity requested. **Date of publication:** 26 July 2023 (online & print)

Citation: Preethi, S.J. & P. Ponmurugan (2023). Developing a fast, reproducible, and simple protocol for virtual lichen herbarium using barcoding and QR code techniques. *Journal of Threatened Taxa* 15(7): 23587–23595. https://doi.org/10.11609/jott.8081.15.7.23587-23595

Funding: This work was supported by the Bharathiar University and Rashtriya Uchchatar Shiksha Abhiyan (RUSA 2.0) - Bharathiar Cancer Theranostics Research Centre, Coimbatore, Tamil Nadu, India.

Competing interests: The authors declare no competing interests.

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Author contributions: The concept plan and manuscript editing and modification were done by the corresponding author Dr. P. Ponmurugan and the manuscript writing were done by the first author Ms. S. Jeya Preethi.

Acknowledgements: The authors thank Bharathiar University and Rashtriya Uchchatar Shiksha Abhiyan (RUSA 2.0) - Bharathiar Cancer Theranostics Research Centre, Coimbatore, Tamil Nadu, India for the support.

INTRODUCTION

A herbarium is a collection of preserved plant specimens, which are repositories to safeguard plant samples of a given area or region and serve as reference materials to build knowledge of biological resources. Virtual or digital herbaria are playing an important role as a large number of plant specimens are digitized and serve as virtual museums and as libraries of information about plants (Primack et al. 2004). A Virtual herbarium is a digitized form of biological specimens, containing a collection of digital images of preserved plants or plant parts which in turn is useful in improving availability of specimens to all users. In addition, storage of more samples in less space without using herbarium sheet, maintenance of original colour, shape and size of samples without microbial attack and odour emission are the salient features of a virtual herbarium (Flannery 2013). The information on botanical collections is made accessible through digitization, database development and the internet through Barcoding Library (BL) and Quick Response (QR) codes in the digital era.

Barcodes are created in response to the requirement of industries to develop a system to capture the product data quickly during the check-out process at supermarkets. They are one-dimensional optical representations, where widths and spacing of parallel lines are translated primarily into numeric data (Law & So 2010). The information in the barcodes are decoded by electronic devices, linked to a database. There are applications available on the internet to decode the barcode information. Similarly, QR codes are the 2-dimensional barcodes used in the trademark for a type of matrix which has gained recognition as an effective tool for product information. These codes connect digital resources to printed text, suggesting the potential to enhance paper-based learning materials (Chen et al. 2011). It can be read from any direction in 360° through position detection located at the three corners (Moisoiu et al. 2014).

University of Washington Herbarium has developed a virtual database of around 72 genera of lichens. US National Science Foundation along with North American Lichen Herbaria created a virtual database containing 2.3 million North American lichen and bryophyte specimens (Lai 2006). African plants initiative scheme was established in 2013 with the aim of digitizing type specimens and making these images available on the website (Patmore 2010). The American plant systematics created a rich website on Lewis and Clark's botanical collections and Linnean Society's website exhibited plant specimens, insects, fish, and shells in digital form (Reveal 2008). But no efforts were undertaken to create QR codes for repository biological specimens for quick access to get the information about repository specimens especially for lichens.

A digital herbarium is useful to improve the access of potential application and diversity of the lichens which in turn is useful to maintain lichen resources in India. In addition, lichens are slow growers and require several years to develop thallus to the length of 1 cm (Ahmadjian 1993). Keeping this in mind, studies were undertaken to create a virtual herbarium for lichens using digital picturization, barcoding and QR code techniques in cloud environment. A simple and rapid protocol was standardized to create virtual herbarium for lichens and subsequently made available online for Lichenologists.

MATERIALS AND METHODS

Collection and identification of lichen samples

Lichen samples were collected from various living and non-living substrates in the Eastern Ghats (Kolli & Yercaud hills) and the Western Ghats (Kodaikanal & Nilgiris hills) of Tamil Nadu, India and were identified by following the standard method of Awasthi (2007). Lichen morphology, anatomy, growth forms, powdery appearance and nature of fruiting bodies embedded on the thallus were critically analysed to identify the lichen communities from genus to species level. Chemical tests (K, C, KC, and PD) were employed to observe the colour reactions on lichen thallus including the existence of lichen secondary metabolites. Lichen thallus were examined for the cortical and medullary chemical compounds by thin layer chromatography method using a suitable solvent system (Orange et al. 2001). The specimens were deposited in the Department of Botany, Bharathiar University, Coimbatore, Tamil Nadu, India as per the conventional method.

Digitization and preparation of Barcoding for lichen specimens

The collected lichen thalli were placed in an Image capturing documentation system fitted with a high resolution digital camera (Precision Co, Ltd, India) to capture the overall images of lichens without any background noise error to minimise the pixel size. Lichen images were also directly photographed using high resolution digital cameras or smartphones with different dimensions on the substratum without disturbing the lichen biodiversity. Lichen images were taken to observe specific parts such as isidia, rhizines, apothesia, soredia etc to understand the digitalized herbarium (https://www.digitallichenbu.in/).

Selected images of lichens with smaller pixel size were transferred to a computer terminal installed with a barcode generator software studio containing RFID label software (TBarCode SDK Activator@) to generate barcodes (Ginni et al. 2022). The barcoding data was generated with individual bars along with numeric numbers without any decimal for lichen specimens such as *Chrysothrix candelaris* (L.) J.R.Laundon (BU/BRL/2022/002), *Leucodermia leucomelos* (L.) Kalb (BU/BRL/2022/022), *Heterodernia flabellate* (fee) D.D.Awasthi BU/BRL/2022/012), *Parmotrema andinum* (Mull.Arg.) Hale (BU/BRL/2022/024), *Parmotrema grayanum* (Hue) Hale (BU/BRL/2022/025), *Parmellinella stuppeum* (Taylor) Hale (BU/BRL/2022/031), and *Ramalina intermedia* (Delise ex Nyl.) Nyl. (BU/BRL/2022/036).

Digitization and preparation of QR codes for lichen specimens

In order to create QR codes for each lichen specimen, QR code generator software studio containing MacOSX.pkg. 10.8+Version: 1.0.3 software was used and then processed digitally so as to read the contents rapidly. Attempts were made to read QR codes in both windows PC and mobile phone devices. If the mobile device did not build in any QR code reader, the user needs to download the right decoder from google play store and install it on to the device. The generated image files as QR codes were used to identify the lichen specimens from genus to species level along with detailed descriptions such as distribution, habit and habitat, family, nature of thallus, reproductive structure, chemistry (colour tests) and secondary metabolites of each lichen sample and were documented (Diazgranados & Funk 2013).

RESULTS

Barcodes and QR codes empowered virtual herbarium for lichens was created wherein, the virtual data was made available online. The virtual data such as digital image, name, and descriptions of lichens were presented. Digital picturization as virtual data for lichen identification can be accessed by the end users of both Windows PCs and smartphone mobile devices online using both barcode and QR code techniques. The end users need to download the right decoder software from google play store and install it on to the device for QR codes. On the other hand, a barcode scanner is necessary to scan the barcode to read the data. The barcodes were generated and displayed in the conventional lichen herbarium in Bharathiar University, Coimbatore, India to get the details of particular lichen specimens. If we scan the barcodes, the binomial name of lichen specimens and their accession numbers are displayed.

Lichen specimens such as *Chrysothrix candelaris* (L.) J.R.Laundon*,* belonging to crustose*, Leucodermia leucomelos* (L.) Kalb*, Heterodernia flabellate* (fee) D.D.Awasthi*, Parmotrema andinum* (Mull.Arg.) Hale*, Parmotrema grayanum* (Hue) Hale*, Parmellinella stuppeum* (Taylor) Hale belonging to foliose and *Ramalina intermedia* (Delise ex Nyl.) Nyl. belonging to fruticose growth forms were identified from genus to species level (Image 1). Different genus and groups of the lichens name was given different code to predict the digital herbarium sample. These repository specimens have been deposited in the lichen herbarium as reference materials as per the conventional method of preparation for future taxonomic studies (Image 2). By using the barcode generator software studio, barcodes were generated and labelled properly for each repository specimen along with their accession numbers (Table 1).

According to Table 2, QR codes were created for all of the chosen lichen species and they provide a brief description that includes information on distribution, habit & habitat, growth forms, name of the family, reproductive structure, chemistry (colour tests), nature of thallus structure, and existence of secondary metabolites. Along with detailed descriptions of each lichen specimen, a digital image is also displayed on the screen. A simple, reliable with reproducible protocol was developed to identify the repository specimens using QR code reader significantly (Image 3). Barcode and QR approaches reveal easy identification and prediction of lichen images very fast with a complete description.

DISCUSSION

Lichenologists identify lichen species routinely by their external and internal morphology along with chemical constituents contained in thallus and to some extent to molecular traits by means of DNA profile (Upreti et al. 2005). Lichen taxonomy is a very complex and time-consuming process that also suffers from shortage of skilled manpower (Nayaka & Upreti 2013). A large number of lichens are being preserved

Table 1. Barcoding for lichen specimens and the accession number.

	Barcode	Binomial name	Accession number
1	9782022002	*Chrysothrix candelaris* (L.) J.R.Laundon	BU/BRL/2022/002
2	9782022022	*Leucodermia leucomelos (L.) Kalb*	BU/BRL/2022/022
3	9782022012	*Heterodernia flabellate* (fee) D.D.Awasthi	BU/BRL/2022/012
4	9782022024	*Parmotrema andinum* (Mull.Arg.) Hale	BU/BRL/2022/024
5	9782022025	*Parmotrema grayanum* (Hue) Hale	BU/BRL/2022/025
6	9782022031	*Parmellinella stuppeum* (Taylor) Hale	BU/BRL/2022/031
7	9782022036	*Ramalina intermedia* (Delise ex Nyl.) Nyl.	BU/BRL/2022/036

using conventional herbarium methods for a variety of research and teaching and learning purposes in the world. But to the best of our knowledge, no attempt has been made so far for developing a virtual herbarium for lichens using digital picturization, barcoding library and QR code techniques in India. The present study was developed to establish a virtual herbarium for lichens with a simple, reliable and user-friendly protocol (Image 3). For a few specimens a barcode library and QR code information virtual data were developed and made available on the website https://www.digitallichenbu.in/ (Tables 1 & 2).

Both barcodes and QR codes showed brief information about the lichen characteristic features in a machine-readable optical label structure. It is used extensively in research for barcoding of flora and fauna in the digital world. Each barcode image is programmed to identify the name of the plant and other information relevant to the plant family, order and taxonomical description. A large number of benefits of QR codes and barcoding system have been listed out (Chase & Fay 2009) like improved inventory management,

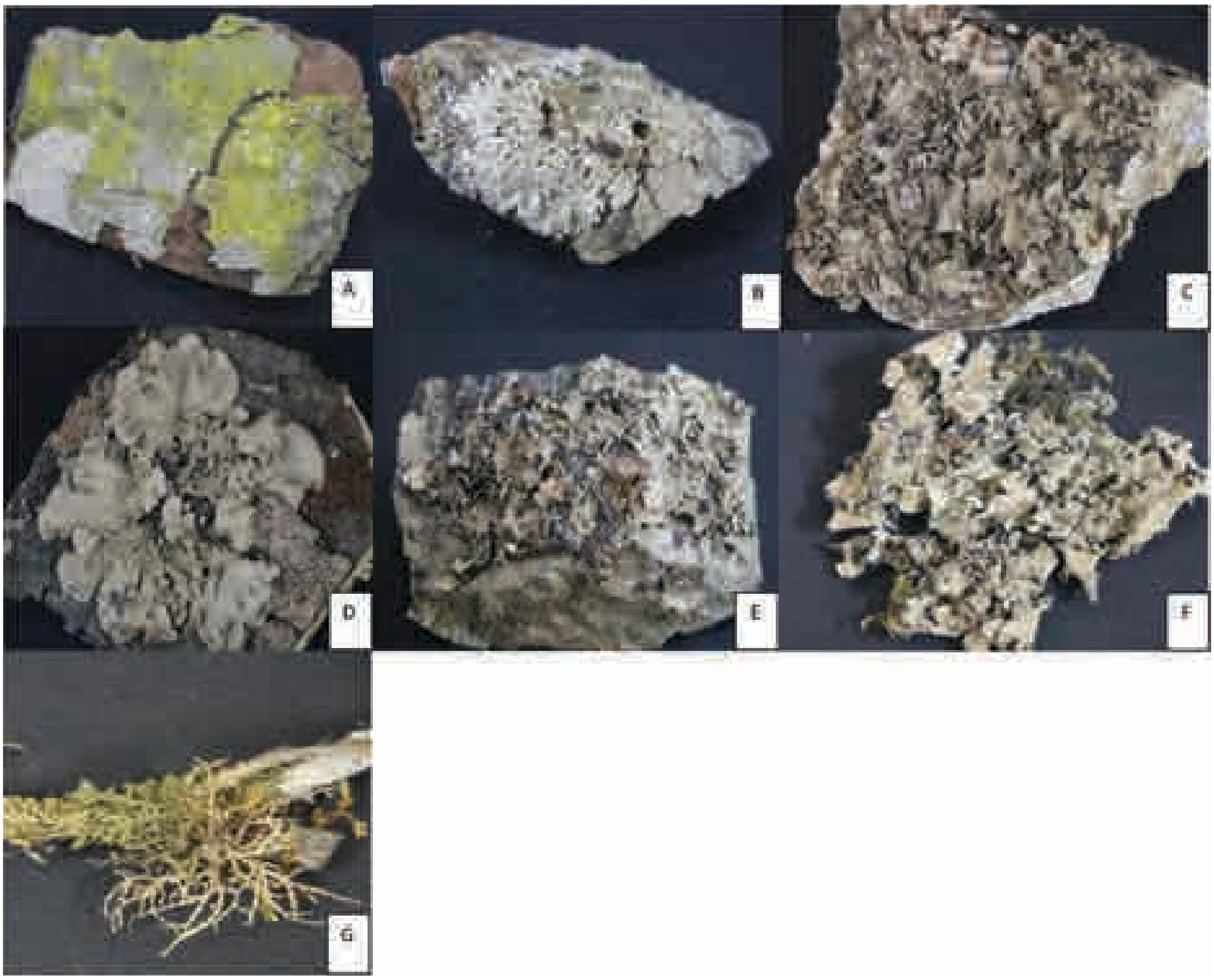

Image 1. Lichen species used for creating lichen virtual herbarium: A—*Chrysothrix candelaris* (L.) J.R.Laundon | B—*Leucodermia leucomelos* (L.) Kalb | C—*Heterodernia flabellate* (fee) D.D.Awasthi | D—*Parmotrema andinum* (Mull.Arg.) Hale | E—*Parmotrema grayanum* (Hue) Hale | F—*Parmellinella stuppeum* (Taylor) Hale | G—*Ramalina intermedia* (Delise ex Nyl.) Nyl. © https://www.digitallichenbu.in/

faster check-in and check-out facility, easy to sort out the specimens, reduced staff workload and skilled man power and increased accuracy and efficiency (Singh 2016).

Virtual herbarium of angio-spermic plants of the Western Ghats of Maharashtra, India, is available with the Modern College of Arts, Science and Commerce, Pune, Maharashtra, India, in which a list of about 1,000 species was made, of which 650 plants were documented and the data on 350 plants is currently available on the website (Singh & Sharma 2009). The primary objective of this project was to capture and store high quality digital images of plant species and to make this database available to students, researchers, and public to disseminate the awareness of regional plants (http://www.indianflora.org/). A virtual herbarium for the higher plants has been created at the Kerala Forest Research Institute, Peechi, Kerala, India, which provides a total of 5,718 records representing 203 plant families and is rendered accessible at http://kfriherbarium.org/ (Sreekumar et al. 2017). Similarly, a digital herbarium for the flora of Karnataka was carried out by Rao et al. (2012) at the Indian Institute of Science, Bangalore, India.

The high-resolution images of digitized plant specimens through virtual herbarium techniques may be useful to examine micro-morphological features of plant parts and can further access the repository specimen information recorded on the data sheet. In addition, using barcodes, plant specimens could be sorted out based on family and order as per the classification with more accuracy and efficiency in the virtual herbarium (Dmitry et al. 2017). It is reported that virtual lichen herbarium is less time-consuming and needs fewer

Table 2. QR codes of lichen specimens and their brief descriptions.

	Lichen species	QR code	Descriptions of lichen species
1.	*Chrysothrix candelaris* (L.) J.R.Laundon		**ID:** BU/BRL/2022/002 **Distribution**: Eastern and Western Ghats of Tamil Nadu, India **Habitat**: Found in Angiosperms tree barks **Ecology**: Open habitats and attached with substratum **Family**: Parmeliaceae **Thallus**: Crustose, leprose, unstratified or, in thick specimens, sometimes indistinctly stratified, indeterminate, thin, irregularly spreading, sometimes forming scattered granules, but usually ±continuous **Upper surface**: Bright yellow throughout, often with an orange or greenish tinge, composed of a mass of fine soredia, 12–30(-40) µm in diam**.** **Medulla**: Usually not evident, in thick thalli sometimes indistinctly present, yellow **Apothecia**: Ascomata, up to 0.5 mm in diam., ±superficial disc: pale orange, often yellow-pruinose margin: thin, ecorticate, soon becoming excluded exciple: poorly developed, composed of anastomosing hyphae epihymenium: hyaline, up to 18 µm tall, composed of of a reticulate layer of richly branched paraphysoids **hymenium:** hyaline, up to 50 µm tall (including epihymenium); paraphysoides: 1–1.5 µm wide, richly intertwined in epihymenium; hypothecium: colorless, poorly developed **Ascospores**: Asci: Clavate, 8-spored ascospores: 9–14 x 3 µm **Pycnidia**: Not observed **Reproductive Structure**: Apothecia **Chemistry**: K- or K+ Orange, sometimes darkening to red-black, C-, KC-, P- or P+ orange; UV+ dull orange or UV- **Secondary metabolites**: calycin and/or pinastric acid
2.	*Leucodermia leucomelos* (L.) Kalb		**ID :** BU/BRL/2022/022 **Distribution**: Eastern and Western Ghats of Tamil Nadu, India **Habitat**: Found in both Gymnosperms and Angiosperms tree barks, rocks and soil **Ecology**: Open habitats and loosely with substratum **Family**: Physciaceae **Thallus**: Foliose type, pendulous and covering large areas, corticolous or terricolous **Lobes**: dichotomously branched, ascending, tapering at apices **Upper surface**: White or cream coloured, sorediate **Lower surface**: Canaliculated, pinkish-brown, erhizinate **Soredia:** Common at apices **Medulla**: White **Apothecia**: Rare, sub terminal, **Ascospores**: Ellipsoid, 8 spores, **Reproductive Structure**: Soredia **Chemistry**: Cortex K^+ yellow, C^-, KC^-, P^+; Medulla K^- or K^+ (yellow to red), C^-, KC^-, P^-or P^+ (red); TLC method detected sekikaic acid, zeorin, chloroatranorin, zorsticitc acid and salazinic acid **Secondary metabolites**: Sekikaic acid, zeorin, chloroatranorin, zorsticitc acid and salazinic acid
3.	*Heterodernia flabellate* (fee) D.D.Awasthi		**ID :** BU/BRL/2022/012 **Distribution**: Eastern and Western Ghats of Tamil Nadu, India **Habitat**: Found in both Gymnosperms and Angiosperms tree barks **Ecology**: Open habitats and loosely with substratum **Family**: Physciaceae **Thallus**: Foliose to sub fruticose, often in loose rosettes or forming tangled mats, loosely adnate or, in part, unattached, 5–15 cm wide **Lobes**: 0.7–2.5 mm wide, ca. 2–4 mm wide at the tips, plane to weakly convex, sublinear to linear-elongate, regularly to irregularly branched, radiating; apices not ascending, contiguous to discrete, with short lateral lobes **Upper surface**: Gray-white to greenish-white, ±partly blackened in the center. **Lower surface**: **Soredia**: lacking Soredia, Isidia, and Pruina **Medulla**: White, lower medulla dark yellow to orange-brown **Apothecia**: Common, laminal, sessile to sub stipitate, 1–6 mm wide; margin crenate at first, lobulate at maturity; inner surface of lobules ecorticate, yellow-orange pigmented; disc concave, dark brown to brown-black, epruinose or weakly white pruinose **Ascospores**: Polyblastidia-type, ellipsoidal, with 2–3 sporoblastidia present at maturity, 27–40 × 12–19 µm. Pycnidia common, immersed, then becoming emergent, visible as black dots; conidia bacilliform, 4–5 × 1 µm **Reproductive Structure**: Apothecia **Chemistry**: Cortex K+ (yellow), C–, KC–, P+ yellow; upper medulla K+ yellow, C–, P–; lower surface K+ violet; **Secondary metabolites**: atranorin (major), zeorin (major), 16β-acetoxyhopane-6α,22- diol (major), leucotylin (minor), 7- chloroemodin (minor), flavoobscurins A, B1, B2 (minor)

	Lichen species	QR code	Descriptions of lichen species
4.	*Parmotrema andinum* (Mull.Arg.) Hale		**ID: BU/BRL/2022/024** **Distribution**: Eastern and Western Ghats of Tamil Nadu, India **Habitat**: Found in both Gymnosperms and Angiosperms tree barks **Locality**: Yercaud hills of Eastern Ghats (Altitude 1515 meters above MSL height, 11.7211° N & 78.1835° E) **Ecology**: Open habitats and loosely with substratum **Family**: Parmeliaceae **Thallus**: Foliose, loosely attached to the substratum. **Lobes**: Lobes ascending, rotund, up to 5–10 mm wide, 120–180 µm thick; margin crenate, eciliate **Upper surface**: Ashy white to grey, smooth, maculate **Lower surface**: Black, slightly wrinkled, with 3–5 mm wide, erhizinate marginal zone. Rhizines in the center, simple, short up to 1mm long **Medulla**: White, 100–120 µm thick **Apothecia**: Rare, Stipitate, up to 10 mm in diameter, disc brown, amphithecium rugose, maculate, epithecium brown, 15–20 µm thick; hymenium 55–65 µm high. Asci clavate, 8-spored, 30–45 x 19 µm **Ascospores:** Spores colourless, simple, ellipsoid, 14–22 x 7–10 µm. Pycnidia laminal, towards apices, black. Conidia filiform, 10–15 µm. Long. **Reproductive Structure**: Apothecia **Chemistry**: Cortex K^+ (yellow), yellow: medulla K-, C+ red, KC + red, P- **Secondary metabolites**: Leconoric acid
5.	*Parmotrema grayanum* (Hue) Hale		**ID:** BU/BRL/2022/025 **Distribution**: Eastern and Western Ghats of Tamil Nadu, India **Habitat**: Found in Angiosperms tree barks and in rocks. **Ecology**: Open habitats and loosely with substratum **Family**: Parmeliaceae **Thallus**: Foliose type, Saxicolous **Lobes**: Rotund to irregular; margins **Upper surface**: Pale grey to grey green, shiny, becoming dull towards the thallus center, somewhat longitudinally folded in the marginal region and emaculate, granular to filiform, simple to coralloid, branched, thin, brown tipped or concolorous **Lower surface**: Black, minutely wrinkled, smooth, shiny, with a broad, erhizinate, pale brown to dark tan marginal zone; rhizines sparse, simple with short. **Soredia**: Abundantly sorediate; soralia marginal, linear to labriform (± crescent-shaped), or subcapitate; soredia ± granular, typically discolored by a dark gray tinge, pale inside **Medulla**: White **Apothecia**: Not seen **Reproductive Structure**: Isidia **Chemistry**: Cortex P+ yellow, K+ yellow, KC–, C–, UV–; medulla P–, K–, KC–, C–, UV– **Secondary metabolites**: Atranorin, and protolichesterinic acid
6.	*Parmellinella stuppeum* (Taylor) Hale		**ID:** BU/BRL/2022/031 **Distribution**: Eastern and Western Ghats of Tamil Nadu, India **Habitat**: Found in both Gymnosperms and Angiosperms tree barks **Ecology**: Open habitats and loosely with substratum **Family**: Parmeliaceae **Thallus**: Foliose, adnate to loosely adnate, 2–20 cm in diam., lobate. **Lobes**: Sub irregular, elongate, slightly imbricate, plane, separate, 4–8 mm wide; apices: rotund, ciliate; cilia: up to 2.0 mm long. **Upper surface**: Gray, smooth, dull, emaculate **Lower surface**: black with brown, naked zone peripherally, centrally rhizinate; rhizines: scattered, simple, black **Soralia**: Granular, common, in linear to orbicular, laminal or marginal soralia **Medulla**: White with continuous algal layer **Apothecia**: Rare, Sub stipitate, up to 30 mm in diam.; margin: crenulate; disc: brown, imperforate. **Ascospores**: Ellipsoid, 12–17 x 6–9 µm Pycnidia: common, punctiform conidia: sublageniform, 4–6 x 1 µm **Reproductive Structure**: Soralia, Ascospores **Chemistry**: Upper Cortex K+ yellow, C-, KC-, P-; medulla K+ yellow turning deep red, C-, KC-, P+ orange. **Secondary metabolites**: atranorin, chloroatranorin, salazinic acid and consalazinic acids (minor).
7.	*Ramalina intermedia* (Delise ex Nyl.) Nyl		**ID**: BU/BRL/2022/036 **Distribution**: Eastern and Western Ghats of Tamil Nadu, India **Habitat**: Found in both Gymnosperms and Angiosperms tree barks **Ecology**: Open habitats and hanging from substratum **Family**: Ramalinaceae **Thallus**: Fruticose, caespitose, up to 3 cm long **Lobes**: Sparingly branched from a narrow holdfast branches: flat, +dorsiventral or subcylindrical, irregular in thickness in cross section, tips often ending in soralia, up to 1.5 mm wide **Upper surface**: Greenish or gray, rarely canaliculated and smooth **Lower surface**: Thin; chondroid strands: continuous, cracked **Soralia**: subterminal or marginal soralia that are 0.4–0.5 mm in diam. **Pseudocyphellae:** Occasional **Medulla**: White **Apothecia**: Not observed **Reproductive Structure**: Soralia **Chemistry:** Cortex K-, C-, KC+ yellow, P-; medulla K-, C-, KC-, P- **Secondary metabolites**: usnic acid, homosekikaic acid, sekikaic acid, 4'-O-methylnorhomosekikaic acid (minor).

Image 2. Conventional lichen herbarium exhibiting barcodes and QR codes.

skilled manpower; also explores the bioactive properties of lichen genera for industrial applications (Flannery 2013). A digital lichen herbarium might be useful to researchers to easily access the lichens of the specific herbaria for their studies.

To conclude, lichen specimens such as *Chrysothrix candelaris* (L.) J.R.Laundon, *Leucodermia leucomelos* (L.) Kalb, *Heterodernia flabellate* (fee) D.D.Awasthi, *Parmotrema andinum* (Mull.Arg.) Hale, *Parmotrema grayanum* (Hue) Hale, *Parmellinella stuppeum* (Taylor) Hale, and *Ramalina intermedia* (Delise ex Nyl.) Nyl. collected from the Eastern Ghats and the Western Ghats of Tamil Nadu, India were digitalized for making a virtual herbarium. The barcodes and quick response (QR) codes were used in the virtual lichen herbarium for quick access and to get a complete description of the repository specimens based on morphological, anatomical and biochemical characterization traits. The present attempt may be highly useful to lichenologists and biodiversity conservation scientists to get information about lichens in digital form without disturbing the lichen biodiversity in the habitats.

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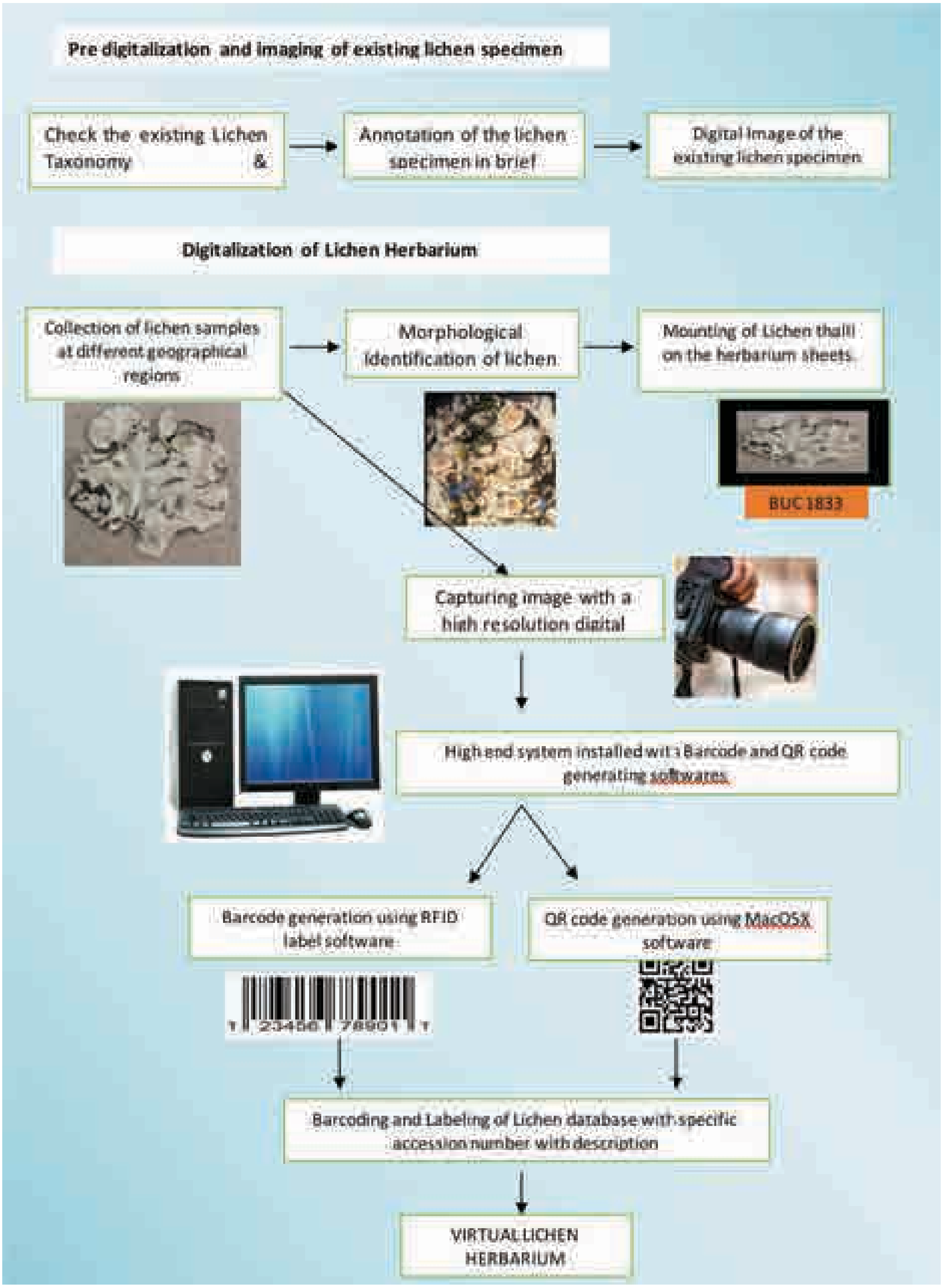

Image 3. Steps involved in creating a virtual herbarium for lichens using barcode and QR code techniques.

Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23596–23600

OPEN ACCESS

ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)

https://doi.org/10.11609/jott.8500.15.7.23596-23600

#8500 | Received 30 April 2023 | Final received 05 July 2023 | Finally accepted 07 July 2023

SHORT COMMUNICATION

Population status of Oriental Darter *Anhinga melanogaster* Pennant, 1769 (Aves: Suliformes: Anhingidae) in Keoladeo National Park, India

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Abstract: Oriental Darter *Anhinga melanogaster* belonging to the family Anhingidae is a globally 'Near Threatened' species occurring in southern and southeastern Asia. The Keoladeo National Park (KNP), Bharatpur, Rajasthan is also known to harbour some population of this species where this study was carried out to assess the population status. Eight wetland blocks were surveyed in different seasons from January 2021 to December 2021 in KNP. The average population of Oriental Darter was found to be maximum (112.8 ±43.8 SE, n = 8) in the winter season, whereas the least (1.8±1.1 SE, n = 8) in the summer season. The maximum population size of darters among the eight wetland blocks was witnessed in Block D of KNP harbouring a mean population of 84.3±20.2 SE (n = 12), whereas the minimum population occurred in Block F (0.3±0.16 SE, n = 12). However, seasonally the total number of darters recorded in all eight wetland blocks during winter, summer and monsoon was 287, 83, and 212, respectively. The findings of the current study reveal that the KNP sustains a viable resident population of Oriental Darters. Further studies are therefore recommended for understanding the seasonal movement pattern and other ecological aspects for its long-term conservation planning.

Keywords: Bharathpur, distribution, Near Threatened, population size, Rajasthan, waterbirds, wetlands.

The Oriental Darter *Anhinga melanogaster* is a relatively large, sleek waterbird, that inhabits shallow inland wetlands, including lakes, rivers, swamps, reservoirs, estuaries, tidal inlets, mangroves, and coastal lagoons. They resemble cormorants and herons in body structure and appearance. They are distributed throughout the oriental region and are resident birds in Pakistan, India, and Sri Lanka. Some populations of the species are also found in other countries including Nepal, Bangladesh, Myanmar, Thailand, Laos, Vietnam, Cambodia, Malaysia, Singapore, Brunei, Indonesia, and Timor-Leste (BirdLife International 2023). In India, darters are widespread, from coastal wetlands to about 300 m in the foothills of the Himalaya, and can also be found at 700 m in Periyar Lake in the Western Ghats (Image 1). Darter has been documented to breed in several locations across India, with KNP and Bhitarkanika Wildlife Sanctuary in Odisha serving as the species' well-known nesting sites (Rahmani 2005). In KNP, Oriental Darter appears to be a local migrant because there is variation in its numbers seasonally. It is an indicator species because its presence in a wetland ecosystem specifies that it holds sufficient fish as prey base. However, darters move away from their natal areas in response to drought conditions. It prefers clear, clean, stagnant water bodies (Kumar et al. 2005). Darters are colonial nesting species and prefer to nest with other waterbird species in the heronry, and move locally

Editor: H. Byju, Coimbatore, Tamil Nadu, India. **Date of publication:** 26 July 2023 (online & print)

Citation: Imtiyaz, N. & S. Kumar (2023). Population status of Oriental Darter *Anhinga melanogaster* Pennant, 1769 (Aves: Suliformes: Anhingidae) in Keoladeo National Park, India. *Journal of Threatened Taxa* 15(7): 23596–23600. https://doi.org/10.11609/jott.8500.15.7.23596-23600

Funding: The first author was awarded Junior Research Fellowship and subsequently Senior Research Fellowship by the University Grants Commission to conduct research.

Competing interests: The authors declare no competing interests.

Acknowledgements: We wish to thank the entire field posted staff of the Forest Department, Keoladeo National Park, in particular the Park Director, Mr Mohit Gupta (DCF) for the continuous help extended by him and equally to his predecessor, Dr. Ajit Uchoi. We are also thankful to the Chief Wildlife Warden, Rajasthan to issue research permit to conduct this study in KNP. All help extended by the Field Assistants Mr. Dinesh Kasna and Mr. Yogesh in the field is duly acknowledged. We are also thankful to one anonymous reviewer for going through the earlier drafts of this paper.

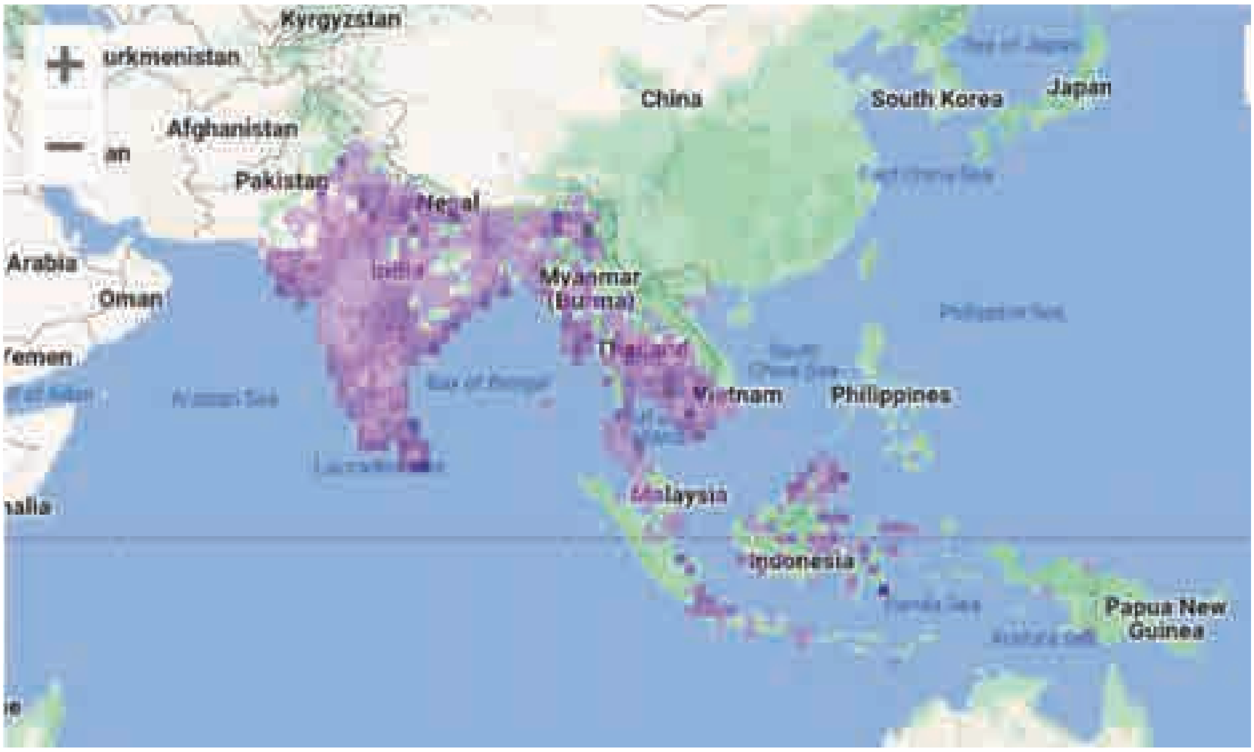

Image 1. Distribution range of the Oriental Darter. Source: www.ebird.org, downloaded on 12 May 2023.

depending on the water conditions (Ali & Ripley 1987; Daniel & Ugra 2003; Kumar et al. 2005).

Due to the decline in the population of the species, it has been listed as 'Near Threatened' by the IUCN Red List of Threatened Species. The population decline has been attributed to pollution, the draining of wetlands, hunting and stealing of eggs and nestlings (BirdLife International 2013). In India, it is legally protected under Schedule II of the Indian Wildlife Protection Act, 1972. There are no current population evaluations of darters from India (BirdLife International 2023). Based on its long-term abundance index over 25 years, current annual trend in abundance over the past five years and the size of its distribution range, its status has been classified as being of low concern in India (SoIB 2020). Keeping in mind the IUCN 'Near Threatened' status of the species, the present study was undertaken in KNP in Rajasthan aiming to estimate the population and the seasonal population fluctuations of the Oriental Darter.

Study Area

KNP is located in Bharatpur district of Rajasthan ($27^0$118'–$27^0$200' N and $77^0$484'–$77^0$552' E) (Figure 1). It is a low-lying area in the floodplains of river Banganga and Gambhir, which are tributaries of river Yamuna covering an area of about 29 km^2. The Park is flat with a gentle slope towards the centre, forming a depression, the total area of which is about 8.5 km^2, which receives migratory waterfowls every year (Vijayan 1987; Ishtiaq 1998). It is a Ramsar site as well as a World Heritage Site identified by UNESCO. The park, known locally as 'Ghana', is a mosaic of dry grassland, woodlands, swamps, and wetlands. The heronry in the park is formed by 15 species of birds, viz., Painted Stork *Mycteria leucocephala*, Asian Openbill *Anastomus oscitans*, Grey Heron *Ardea cinerea*, Purple Heron *Ardea purpurea*, Black-crowned Night Heron *Nycticorax nycticorax*, Great Egret *Ardea alba*, Intermediate Egret *Ardea intermedia*, Little Egret *Egretta garzetta*, Cattle Egret *Bubulcus ibis*, Black-headed Ibis *Threskiornis melanocephalus*, Little Cormorant *Microcarbo niger*, Indian Cormorant *Phalacrocorax fuscicollis*, Great Cormorant *Phalacrocorax carbo*, Oriental Darter *Anhinga melanogaster*, and Eurasian Spoonbill *Platalea leucorodia*.

Methods

The total count method (Bibby et al. 2000) was employed to assess the population status of darters in the study area. The Park management has divided the whole wetland into eight blocks (B, D, E, F, K, L, N, G) and we adopted these blocks as such for our surveys (Image

Figure 1. Block wise map of Keoladeo National Park. Source: Verma & Prakash 2007.

1). Data were collected in such a way that all three major seasons were covered, viz.: summer (March–June), monsoon (July–October), and winter (November–February). Darters were counted in each block using 8 x 32 binoculars (Bibby et al. 2000). Simultaneous counts were made on fortnightly basis from elevated points by two observers in each block from 0060–0080 h during summer and monsoon seasons and from 0080–1000 h during winter season due to fog in early morning hours. Precautions such as reaching early in the park and sensitizing and briefing tourists were exercised to avoid disturbance to darters during counts.

Results

Oriental darters are mainly piscivorous birds that occur singly, or in flocks of small size (usually 3–6 individuals). However, rarely large flocks of up to 20 birds were seen (n = 6). Although several piscivorous waterbirds co-exist with Oriental Darters in KNP, their major competitors with similar food habits of diving and capturing fish are Great Cormorant *Phalacrocorax carbo*, Indian Cormorant *Phalacrocorax fuscicollis*, and Little Cormorant *Microcarbo niger*. It was observed that darters were distributed heterogeneously within the wetland sites, largely in response to the availability of water, and lack of submerged vegetation. Overall, the mean population size of darters was found to be maximum (84.3±20.2, n = 12) in 'block D' and minimum (0.3±0.16, n = 12) in 'block F' of KNP (Table 1).

Total number of darters recorded in all eight blocks during winter, summer and monsoon were 287, 83, and 212, respectively (Table 2). Among all the wetland blocks in KNP, the highest population was recorded in block D in all the seasons, namely winter (195), summer (49) and monsoon (176). On the other hand, the lowest population of darters was recorded in block F in all the seasons, i.e., one individual in winter and none in summer (Table 2).

Discussion

Oriental Darters were found to be territorial in their foraging grounds as they were quite aggressive towards conspecifics whenever they attempted to come close. The distribution pattern of darters was not uniform in the wetland. Such a pattern of distribution can be attributed to the availability of food and the appropriate water depth preferred by these piscivorous birds.

Population counts of Oriental Darter in KNP indicate distinct variation in its population size (Table 2). The average population of the species was highest in winter followed by monsoon and the population of the bird inhabiting the wetland was lowest in the summer season. This may be because in winter, the wetland area is full of water and the prey species of the bird are abundant.

Table 1. Mean population of the Oriental Darter in Keoladeo National Park in different seasons (2021).

Wetland block	Wetland area (in ha)	Season-wise mean population ± SE			Overall mean population (± SE)
		Winter	Summer	Monsoon	
B	1.31	9.0±0.7	3±1.9	33±1.9	15.0±3.9
D	1.38	112.8±43.8	28±7.9	121±34.3	84.3±20.2
E	1.55	10.5±2.1	1.8±1.1	NS	6.1±1.9
F	3.06	0.5±0.2	0.0±0.0	NS	0.3±0.16
K	2.28	14.3±0.7	4.3±1.4	NS	9.3±2.0
L	4.5	29.2±6.8	2.8±1.8	NS	16±5.9
N	0.65	4.8±0.4	1.0±1.0	NS	2.8±0.8
G	0.67	2.0±0.0	0.2±0.2	NS	1.1±0.3

NS—Not surveyed

Table 2. Maximum and minimum population of Oriental Darter during different seasons in Keoladeo National Park (2021).

Wetland block	Winter		Summer		Monsoon	
	Max	Min	Max	Min	Max	Min
B	11	8	8	4	36	28
D	**195**	34	**49**	12	**176**	34
E	15	7	5	**0**	NS	NS
F	**1**	0	**0**	0	NS	NS
K	15	12	8	2	NS	NS
L	42	16	8	0	NS	NS
N	6	4	4	0	NS	NS
G	2	2	1	0	NS	NS
Total	287	83	83	18	212	62

NS—Not surveyed

The mean population of darters was relatively low in blocks G and F because they were less suitable for them due to more submerged vegetation. Also, the post-monsoon period in the park coincides with the post-fledging period of darters, which are therefore seen in higher numbers in winter after the completion of their breeding season.

The Oriental Darter is an obligate piscivore that prefers to forage in shallow waters. However, the African Darters *Anhinga melanogaster rufa* have been reported to dive in waters <5 m deep (Ryan 2007). In summer, water is available only in some of the blocks in the park where darters can be found feeding. Among different sites in the wetland area of the park, Block D was found to support the maximum population of Oriental Darter, which may be due to abundant prey in the deep-water system of the block and partly it can also be correlated with the presence of less submerged and emergent grass species, thereby offering less hindrance to the foraging darters. Most authors have suggested that food resources have been found to influence the distribution and selection of specific habitat types by animals (Johnson 2000; Johnson & Sherry 2001; Narasimmarajan et al. 2012). Furthermore, our results are in accordance with the study of Hustler (1992) who asserted that while diving, African Darters maintained their buoyancy at 2–4 m depth though they may utilize the whole water column. The findings of the current study reveal that the wetland sustains a viable resident population of Oriental Darter in the park. The study further provides information on seasonal variation in its population in the KNP. Ten individuals of Oriental Darter were colour banded during this study but the recapture rate or resighting of the marked individuals was extremely low both within the park and the satellite wetlands in its surroundings. On an average, one individual out of the 10 marked darters was resighted in a month. A large sample size of Oriental Darter is thus required for colour banding to have better recapture rate. In order to understand the dynamics of movement pattern of darters whether they are local migrant in KNP or distant migrants, a conventional radio-telemetry or satellite telemetry or colour banding a large sample size or adequate numbers of darters may be more useful to validate it.

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Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23601–23605
ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)
https://doi.org/10.11609/jott.8607.15.7.23601-23605
#8607 | Received 21 June 2023 | Final received 06 July 2023 | Finally accepted 10 July 2023

SHORT COMMUNICATION

Breeding of Himalayan Vulture *Gyps himalayensis* Hume, 1869 (Aves: Accipitriformes: Accipitridae) in the Assam State Zoo, Guwahati, Assam, India

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Abstract: Himalayan Vulture *Gyps himalayensis* has been bred successfully at the Assam State Zoo, Guwahati in 2022. This is the first record of captive breeding of the species in India. The adults were kept in a display aviary in the Zoo where they constructed a nest on ground and laid an egg. The nestling was hand reared in temperature and humidity-controlled boxes and air-conditioned room. It was fed on goat meat and bone pieces and the consumption records maintained. The records of weight gain and body growth were maintained. It took about five months to fledge out.

Keywords: Captive breeding, food consumption, hand rearing, high altitude species, vulture nestling, weight gain.

Himalayan Vulture *Gyps himalayensis* is the largest vulture among the old-world vultures. Its distribution covers a wide range that includes the countries Afghanistan, Bhutan, China, India, Kazakhstan, Kyrgyzstan, Mongolia, Nepal, Pakistan, Tajikistan, and Uzbekistan. In India, this species breeds in the Himalaya and in winter the immature population visits the plains (Ali & Ripley 1983). The species is categorised as 'Near Threatened' considering that about 66,000–334,000 individuals exist in the wild (Botha et al. 2017). The species is a common winter visitor to plains of India that includes Assam. This migration is done only by the immature birds in winter while the adults remain in the breeding ground. The Himalayan Vultures feed on livestock carcasses along with the local resident *Gyps* vultures – the White-rumped Vulture *Gyps bengalensis* and the Slender-billed Vulture *Gyps tenuirostris*. Their feeding habit makes them vulnerable to the unintentional poisoning (Botha et al. 2017). As the feral dogs attack the livestock, in retaliation, the poison baits are used by the cattle owners to kill the dogs and wild carnivores. The vultures get attracted towards these poisoned baits and feed on them. The victimized vultures in such incidences were rescued and saved by the team of the Bombay Natural History Society and Forest Department in Assam. The Himalayan Vultures saved from such incidences in 2011–12 were kept in the display of the Assam State Zoo and Botanical Park at Guwahati.

Housing and diet in the Zoo

Ten Himalayan vultures were kept for display in a 40 x 30 ft aviary, covered with galvanized mesh. The

Editor: H. Byju, Coimbatore, Tamil Nadu, India. **Date of publication:** 26 July 2023 (online & print)

Citation: Ranade, S., J. Gore & A. Kumar (2023). Breeding of Himalayan Vulture *Gyps himalayensis* Hume, 1869 (Aves: Accipitriformes: Accipitridae) in the Assam State Zoo, Guwahati, Assam, India. *Journal of Threatened Taxa* 15(7): 23601–23605. https://doi.org/10.11609/jott.8607.15.7.23601-23605

Funding: None.

Competing interests: The authors declare no competing interests.

Acknowledgements: The authors are thankful to the Central Zoo Authority and the Bombay Natural History Society, India. The authors express their gratitude towards Mr. M.K. Yadava, PCCF WL & CWLW, Dr. Amit Sahai PCCF & HOFF, Assam, and Mr. Tejas Mariswami, former director, Assam Zoo. The authors are thankful to the veterinary team Dr. Shankar Sharma, Dr. Parikshit Kakati, Dr. Percy Avari, Dr. Panchami Sharma and Dr. Indrani Borgohain, range officer. The hard work by all staff of the Assam State Zoo and the staff of Vulture Conservation Breeding Center, Rani is acknowledged.

vultures were provided with meat daily. Water trough and perches were provided for vultures. The vultures took more than five years to mature and one pair was formed in the flock. The mating pair did not prefer the nest ledge that was provided to them. Instead, they constructed a nest in a secluded corner of aviary on ground when pine twigs were made available to the pair during the months of December and January. In the first two years (2019–20), the parents failed to construct a nest and incubate the egg, and the egg perished. In 2021, a hatching problem was noted when the chick was stuck at the piping stage. It is the stage at which the developed embryo breaks the shell with an egg-tooth on its upper mandible and try to come out. The hatchling was rescued that survived for six days but died due to unknown reason. The systematic post-mortem could not provide any clue and tests for bacteriology and virology were negative. In 2022, the successful hatching was noted on 14 March and the nestling was shifted to the artificial brooding facility on 15 March.

Housing for nestling

During first month, the nestling was kept in the brooder made up of a plastic box (1 x 1 x ½ ft) with a mat for the grip. The temperature was maintained around 30–35° C with a lamp, a water bowl and it was monitored with a thermo-hygrometer. The nestling was provided with sufficient space to move towards and away from the heat source.

As the nestling grew up in size during the second, third and the fourth months, the nestling was transferred to larger boxes successively. The room temperature and humidity were maintained with air conditioner and de-humidifiers.

During the fifth and the sixth month, the nestling was kept in a temperature and humidity-controlled room, on an artificial nest. The nest consisted of a layer of leaves that would soak up excreta. Perches were provided on all four sides that not only avoided the accidental fall of the nestling but also encouraged the nestling to perch on it.

Food for nestling

The nestling started to feed from the second day after hatching (16 March 2022). The nestling was fed with very small pieces of goat meat in the first week and the food quantity was increased as the days passed. From seventh day onwards, the nestling was fed on pieces of ribs as Calcium supplement. From one month onwards, the nestling was provided with goat tail that contained bones. In addition, the nestling was fed with small pieces of muscles, liver and skin. In the beginning, the nestling digested the bones pieces completely but on day 138, it regurgitated bone pieces and hairs in casts for the first time. Taking it as a cue, the daily feeding of additional bone piece was stopped though the goat tail was fed till end of the sixth month.

Frequency of feed

In the first month, feeding was carried out six times a day, from second to fifth month, feeding was done four times a day while in the sixth month it was twice a day. The average fortnightly food consumed by the nestling is represented in the graph. The graph shows steep hike in food consumption in first three months—April, May, and June. After that the graph rises gradually, forming a plateau from September onwards. As the bird had fledged out in mid-August, by that time it was almost completely developed. In the next couple of months, only the primaries and tail grew up to the fullest. It could the reason of decreased appetite of the fledgling in October, but it resumed again once the energy consuming flight exercise was added to the daily routine (Figure 1).

Growth of nestling

The weight of the nestling was recorded with a digital weighing balance. The nestling growth took place somewhat exponentially till it fledged out in August. Afterward, the bird gained weight gradually in next two months and stabilized at 7 kg. In nature, the juveniles of Himalayan Vulture migrate to plains in November and must be evolutionary programmed to gain weight as the energy reserve for the purpose (Figure 2). The periodic photographic record was maintained to understand the development of the plumage (Image 1–6).

A few important physical and behavioural milestones achieved during the nestling phase:

Day	Important event
0	Successful hatching took place on 14 March 2022
2	The nestling opened its eyes fully
5	The nestling showed first attempt to preen itself
8	It quivered the wings to grab attention for feeding
10	The nestling was able to feed itself from a bowl. Its downy coat became dense
19	Its claws began to harden
50	Interscapular, humeral and wing coverts started to appear as the brush
90	Primaries started to appear as the brush
107	It showed reaction to its own image by hissing at it
111	The nestling opened its wing for sunning
120	The nestling was introduced to ground with grass for half an hour.
150	Primaries developed completely, though tail was a bit short

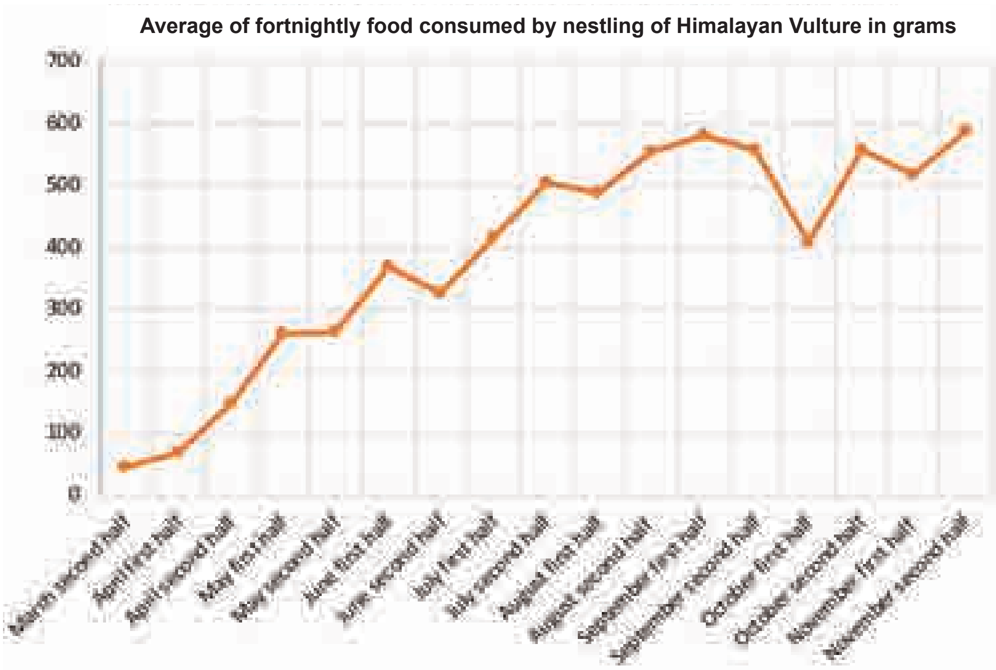

Figure 1. Average fortnightly food consumed by the hand reared nestling of Himalayan Vulture at Assam, India 2022.

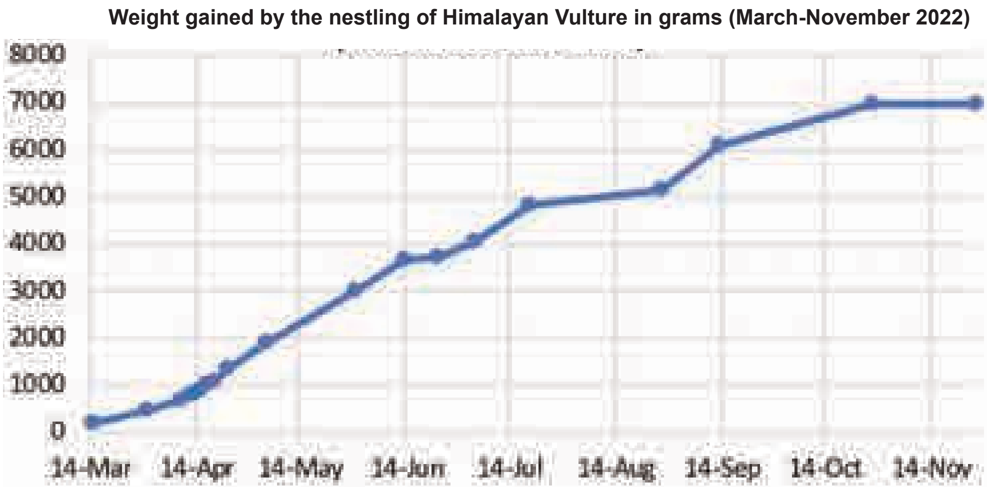

Figure 2. Weight gained by the nestling of Himalayan Vulture.

Colouration of nestling

In the first week, the skin around the eyes of the nestling was grey coloured. The skin around tail portion was also grey in colour. The cere and legs of the nestling were pink while rest of its body was covered in whitish downy. Till one and a half month, the nestling appeared whitish after which the coverts grew very fast. The coverts were chocolate brown coloured with an off-white streak along the rachis. The primary and secondary outer coverts on wings had an off-white blotch at terminal position. The overall body of the nestling started to appear chocolate brown in colour which is a typical coloration of the juvenile. The primaries grew up by the end of fifth month (Image 1–6). The morphometric records were taken on the day 160, when the bird attempted to leave the nest-ledge and jumped out. The morphometrics recorded were as follows: beak 50 mm, cere 30 mm (depth 32 mm), tarsus 120 mm (width of tarsus 14 mm and height of tarsus 17 mm), wing cord 680 mm and tail 380 mm. Even after the event of jumping out of the nest-ledge, the fledgling continued to stay on the nest or remained nearby the

Image 1. Nestling with white downy coat in first week (Day 6)

Image 2. The coverts started to appear as brownish colored brush tips among the thick downy feathers (Day 67).

Image 3. Coverts growing on interscapular tract, humeral tract and wings (Day 82).

Image 4. Well developed coverts, while the primaries and tail begun to grow (Day 130).

Image 5. Fledgling in the nest with well-developed coverts and primaries.

Image 6. The juvenile Himalayan Vulture (Day 293).

nest in temperature-controlled room. The nestling was kept in the natural environment in an aviary from day 190 onwards.

Discussion

The Himalayan Vulture is a common winter migrant in Indian plains and resident of the high Himalayas, yet never kept in any zoo for breeding purpose. The Assam State Zoo has a record of keeping a few Himalayan Vultures for display, although all the birds were rescued ones. Till the end of 20th Century, vultures were quite a common sight in the wilderness and very few of them were appreciated, kept in zoos and bred in captivity. Schlee (1989) recorded the first successful breeding of the Himalayan Vulture in the menagerie in Paris. A few more examples of vulture species being hand-reared are Ruppell's Griffon Vulture *Gyps rueppelli* (Schlee 1998), breeding of White-rumped Vulture *Gyps bengalensis* (Sarker & Iqbal 1997), husbandry of Cinereous Vulture *Aegypius monachus* in the North American Zoos (Diebold & White 1989), captive breeding of Lappet-faced Vulture *Torgos tracheliotus* (Mendelssohn & Marder 1983; Beall 1992), breeding of Eurasian Griffon *Gyps fulvus* (Gardener 1980), breeding of Bearded Vulture *Gypaetus barbatus* (Zwart et al. 1991) and rearing of Andean Condor *Vultur gryphus,* and King Vulture *Sarcoramphus papa* (Zwart & Louwman 1978). In India, the Bombay Natural History Society has bred the three species of vultures- White-rumped Vulture *Gyps bengalensis*, Indian Vulture *Gyps indicus* and Slender-billed Vulture *Gyps tenuirostris* in captivity for the conservation and reintroduction purpose (Bowden et al 2012)

The Himalayan Vulture being a high-altitude bird, it is not usual for the species to breed in the low land with tropical and humid climate. Yet, like many mammals and birds, the species acclimatized and managed to breed (Lague 2017).

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Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23606–23609

OPEN ACCESS

ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)

https://doi.org/10.11609/jott.8533.15.7.23606-23609

#8533 | Received 18 May 2023 | Final received 02 July 2023 | Finally accepted 10 July 2023

NOTE

Unusual foraging behaviour of the Bengal Slow Loris *Nycticebus bengalensis* (Lacépède, 1800) (Mammalia: Primates: Lorisidae) in the Shan Highlands, Myanmar

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The Bengal Slow Loris *Nycticebus bengalensis* is a nocturnal, arboreal, and slow-moving animal, which is native to southern and southeastern Asia. It has the largest geographical range of the four species of the genus *Nycticebus* (Rogers & Nekaris 2011; Nijman 2015; Oliver et al. 2019; Nekaris et al. 2020), comprising Bangladesh, Bhutan, northeastern India, China, Myanmar, Thailand, Cambodia, and Vietnam. Found in evergreen, semi-evergreen and deciduous forest, and degraded areas (Rajamani et al. 2009; Swapna et al. 2009; Das et al. 2014; Francis 2019), it frequents large, tall trees with dense foliage canopy (Pliosungnoen et al. 2010), forest edges, and human-modified landscapes, including heavily disturbed areas, such as home gardens (Das et al. 2014, 2016; Kumar et al. 2014). It can move through thick grass along the ground when tree canopy is lacking (Starr et al. 2010; Rogers & Nekaris 2011).

The diet of *Nycticebus bengalensis* includes plant exudates (gum), bark, leaves, nectar, fruit, small invertebrates, and birds' eggs (Swapna et al. 2009; Rogers & Nekaris 2011; Das et al. 2014; Oliver et al. 2019; Nekaris et al. 2020). It lives singly, in pairs, or in family groups (Ankel-Simons 2006; Al-Razi et al. 2020). Currently categorized as Endangered, on account of habitat loss and over hunting (Nekaris et al. 2020), in Myanmar it is theoretically fully protected by the Protection of Biodiversity and Conservation Areas Law, 2018. However, live Bengal Slow Lorises and their parts are extensively traded on the Sino-Myanmar border, especially in Mong La, eastern Shan State, where, according to Nijman et al. (2014), 'thousands of individuals are killed annually to supply the demand from this one market alone'.

Although considered to be widely distributed in Myanmar (Francis 2019; Nekaris et al. 2020), there is little information about the in-country distribution, ecology, and behaviour of this cryptic primate because of its nocturnal lifestyle and arboreal habits. Yin (1993) includes historical records from Tanintharyi Region (Myeik = Mergui, KadanKyun = King Island), Mon State (Kyaikkhami = Amherst), Kayin State (Thandaung), Rakhine State (no data), Sagaing Region (Kindat) and Kachin State (Bhamo, Hai Bum, Singaling Hkamti). Nijman (2015) includes two individuals from near Saw Law in Kachin State and reports that they are rarely seen in

Editor: Mewa Singh, University of Mysore, Mysuru, India. **Date of publication:** 26 July 2023 (online & print)

Citation: Oo, S.S.L., K.A.N. Oo & P.J.J. Bates (2023). Unusual foraging behaviour of the Bengal Slow Loris *Nycticebus bengalensis* (Lacépède, 1800) (Mammalia: Primates: Lorisidae) in the Shan Highlands, Myanmar. *Journal of Threatened Taxa* 15(7): 23606–23609. https://doi.org/10.11609/jott.8533.15.7.23606-23609

Funding: Financial support for field visits was provided by Sai Sai Peanut Oil.

Competing interests: The authors declare no competing interests.

Acknowledgements: We thank Nay Myo Shwe for reviewing the first draft. Our thanks also go to Sai Aung Tun Thein for sharing his photo.

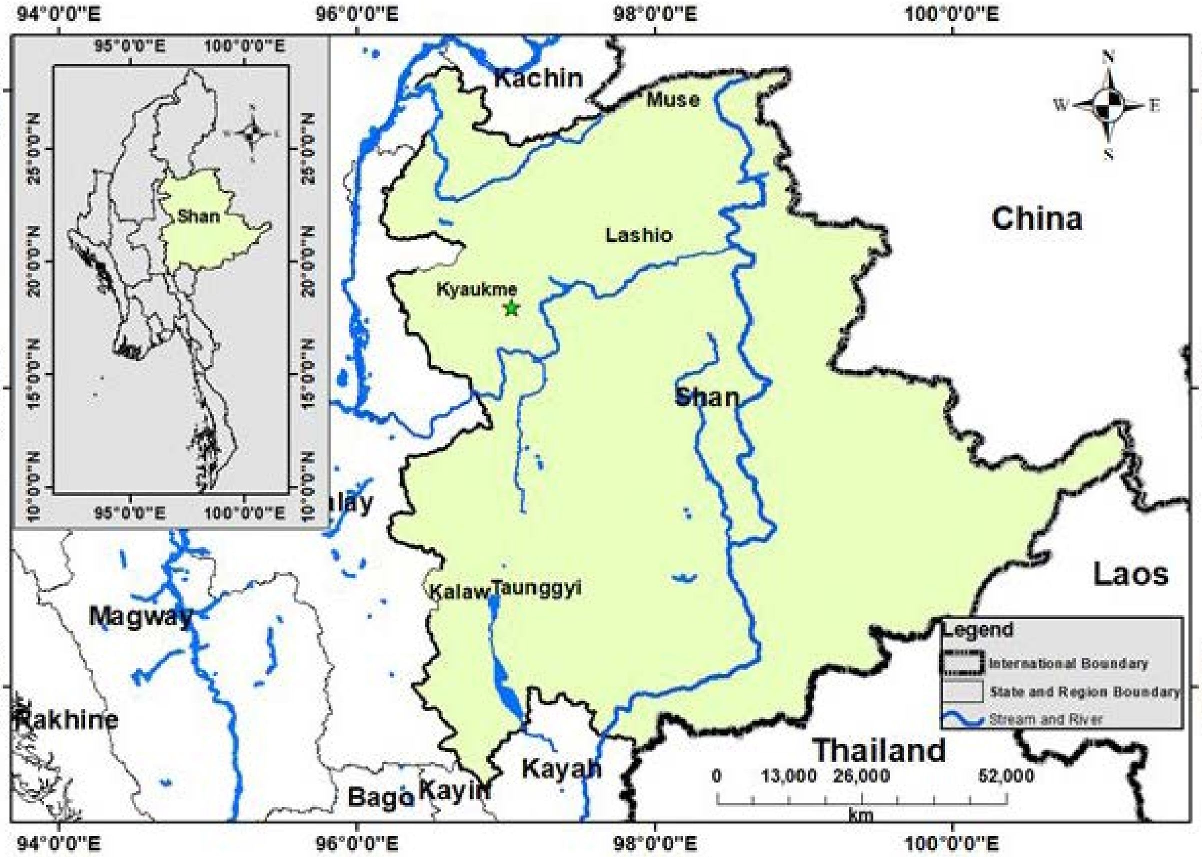

Figure 1. Kyaukme (green star), northern Shan State, where the Bengal Slow Loris was recorded.

this area. On 21 May 2019, a Slow Loris was observed in degraded forest, at an elevation of 1,100 m in Ywangan Township (21.2219°N, 96.5578°E) in southern Shan State.

Recently, a single *Nycticebus bengalensis* was seen entering a house and climbing up to a roof beam where it found nest of a Eurasian Tree Sparrow *Passer montanus*. This happened on 30 April 2022, at 2230 h, in a suburban area of Kyaukme, northern Shan State, Myanmar (22.5489°N, 97.0397°E) (Figure 1). The male slow loris captured one sparrow, which it devoured for about 45 min. It sat on a beam, and after first biting the bird's head, it fed on its prey slowly (Image 1). Although it ate the bones of the bird, it removed most, but not all of the feathers, which fell to the floor (similar observations were made for the Javan Slow Loris *Nycticebus javanicus* in Indonesia (Cabana et al. 2017)). Subsequently, the Loris captured another sparrow and fed again. It was observed that at times it stayed upside down, with its hind feet clinging to the beams of the house, whilst eating its prey. It appeared to be either unaware of the presence of humans or not afraid, as the animal was photographed from within 5 m.

After feeding, the animal climbed down slowly and started to leave the house at 2335 h (30 April 2022) (Image 2). It exited on the electric service line at 0050 h (1 May 2022) (Image 3). Unfortunately, it was not possible to follow this nocturnal animal and record its sleeping quarters as there was a night-time curfew (from 2000–0500 h) in the region after the February 2021 coup. As in Bangladesh (Al-Razi et al. 2019), there is a report (Wildlife of Myanmar, 2022) of a Bengal Slow Loris being electrocuted on a power line in Banmaw (Bhamo), Kachin State, Myanmar.

Situated at an elevation of approximately 780 m, the climate of Kyaukme is humid subtropical with an annual rainfall of about 2,100 mm and a mean annual temperature of 28.9°C. The urban area of Kyaukme is approximately 9.5 km^2, with a population of some 46,000 individuals (General Administration Department 2019). The house where the slow loris was observed is located within a highly disturbed, anthropogenically modified, mosaic of habitats. There is a bamboo grove and a small wood within 100 m and 200 m, respectively, of the house. In Indonesia, the presence of bamboo in human-modified environments has proved important for Javan Slow Loris conservation as it provides essential sleep sites (Nekaris

Image 1. The Bengal Slow Loris feeding on a Eurasian Tree Sparrow within the house. © SSLO

Image 2. The Bengal Slow Loris showing its prominent midline stripe as it climbs down before leaving the house. © SSLO

Image 3. The Bengal Slow Loris moving away from the house on the electricity supply cable. © Sai Aung Tun Thein

et al. 2017). It is noteworthy that Kyaukme is situated en route to the Sino-Myanmar border, with its active wildlife markets and a high demand for wild animals.

This recent observation provides surprising new information about the diet of *Nycticebus bengalensis* and its habituation to highly disturbed urban environments. It suggests that in Myanmar, as elsewhere, urban habitats can serve as a refuge for endangered species (Becker & Buchholz 2016). To understand better its local and national conservation needs, we recommend more detailed studies of the Bengal Slow Loris's ecology, especially habitat preferences, population status and behaviour.

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Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23610–23614

OPEN ACCESS

ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)

https://doi.org/10.11609/jott.8460.15.7.23610-23614

#8460 | Received 29 March 2023 | Final received 08 May 2023 | Finally accepted 05 July 2023

NOTE

Powerline pylons: an unusual nesting success of White-bellied Sea-Eagle *Haliaeetus leucogaster* (Gmelin, 1788) (Aves: Accipitriformes: Accipitridae) from Ramanathapuram, southeastern coast of India

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The White-bellied Sea-Eagle (WBSE) *Haliaeetus leucogaster* (Gmelin, 1788) is a resident raptor belonging to the family Accipitridae. It has a wide distribution range on the sea coast of India from about Mumbai, south to the eastern coast of Bangladesh, and Sri Lanka in southern Asia (del Hoyo et al. 1994), through all coastal southeastern Asia, including Burma, Thailand, Malaysia, Indonesia, Indochina, the main and offshore islands of the Philippines, and southern China, including Hong Kong, Hainan, and Fuzhou, eastwards through New Guinea & the Bismarck Archipelago, and Australia. In the northern Solomons, they are restricted to the Nissan Island (Strange 2000; Ferguson-Lees et al. 2001). According to the IUCN Red List, it is categorized as 'Least Concern' (IUCN 2022).

The WBSE is occasionally seen in inland waters along tidal rivers and in freshwater lakes (Ali & Ripley 1987). It feeds mainly on sea snakes and fish. WBSE builds nests near the seacoast, tidal creeks, and estuaries. This diurnal monogamous bird of prey occupies the same localities for several years in succession and nests in tall trees (Ali 1996). Nesting of WBSE is reported from trees like Mango *Mangifera indica, Casuarina equisetifolia*, Banyan *Ficus bengalensis*, Fig *Ficus religiosa*, Coconut Palm *Cocos nucifera*, Tamarind *Tamarindus indica*, *Sterculia foetida*, *Terminalia paniculata*, Devil's Tree *Alstonia scholaris*, and Baheda *Terminalia bellirica* (Ali 1996; Neema et al. 2021). On the eastern coast of India, nesting in trees are recorded at Bhitarkanika (Gopi & Pandav 2006; Palei et al. 2014), Chilika Lake, and Konark Balukhanda Wildlife Sanctuary (Rahmani & Nair 2012). On the western coast of India, tree nesting is recorded from Raigad, Ratnagiri, and Sindhudurg districts of Maharashtra (Katdare & Mone 2003; Katdare et al. 2004), and the Netrani Islands of Karnataka (Pande et al. 2011).

Observations

The nesting observations were conducted from November 2022 to March 2023. We used binoculars and Canon DSLR cameras with telephoto lenses for observation and pictures. The visual surveys were carried out for recording parameters like: (1) the height of the power line tower, (2) height of the nest from the ground, (3) the width of the artificial structures, and (4) the

Editor: Anonymity requested.

Date of publication: 26 July 2023 (online & print)

Citation: Byju, H., N. Raveendran & A.J. Mathiyazhagan (2023). Powerline pylons: an unusual nesting success of White-bellied Sea-Eagle *Haliaeetus leucogaster* (Gmelin, 1788) (Aves: Accipitriformes: Accipitridae) from Ramanathapuram, southeastern coast of India. *Journal of Threatened Taxa* 15(7): 23610–23614. https://doi.org/10.11609/jott.8460.15.7.23610-23614

Funding: None.

Competing interests: The authors declare no competing interests.

Acknowledgements: Our sincere gratitude to Somu Prasad and his team of Keelakarai Range of Ramanathapuram forest division for logistical support during our field visits. We also thank Kishore. R for helping with the map.

Figure 1. Map showing the nesting site of White-bellied Sea-Eagle from Ramanathapuram district on power pylon.

distance from the sea (Azman et al. 2013).

On 24 November 2022, during one of our routine shorebirds monitoring studies in Ramanathapuram, we observed a large nest on a powerline pylon near the rainwater storage area of Pudumadam (9.289035° N, 78.998988° E) (Figure 1). This storage area was nearly full last year and almost dried up this year. As we passed the first pylon to the next one, we saw another nest of similar size, which made us stop and check the nest from a better position. We observed the presence of WBSEs sitting on the edge of the nest on the first pylon. As we scanned the adjacent pylons, we also found a third nest on the third pylon. Each pylon was at a distance of 100 m from the other (Image 1). These pylons were on the paddy fields adjacent to the rainwater storage area. WBSEs are reported to nest on power poles and transmission towers in Australia and Thailand by birdwatchers. In India, WBSE nesting on a telecommunications tower was reported from Andhra Pradesh (Narayana & Rao 2019).

Image 1. Multiple nests in different pylons constructed by the White-bellied Sea-Eagle.

The height of the nest in the pylon was approximately 18 m (60 ft). The base width of the pylon structure was 180 cm (6 ft). The nest was about 145 m (4 ft) wide (Image 2). The nest is a large deep bowl constructed of thick sticks, twigs, and branches and lined with materials such as grass, seaweed, or green leaves (Image 3). The nesting location was at approximately 2 km aerially from the sea. We maintained a safe distance of about 100 m on the first observation day. Then one adult bird moved away from the nest in the evening. One stayed back in the nest, and the other did not return till dark.

On our subsequent visits on 24–26 December, we observed an incubating adult on the nest on the first pylon. We also found a fourth nest on another pylon (the fourth one) which was absent during the previous observations. Only one nest among the four was utilized by the WBSEs for incubation. False nesting among WBSEs is not reported elsewhere, so this could either be a false nesting since the fourth nest was found during the later observations or the abandoned nests of previous years. The adult male usually visited the nest during the sunrise. As soon as this happened, the incubating adult bird slowly got up and stretched its wings and started flying and soared for about an hour, either alone or with the other adult bird, and returned to the nest. On 6–7 January 2023, we noticed the incubation by an adult bird, and the other adult was not seen till evening. The male bird while reaching the nest, stayed on the edges of the large nest, while the female continued to incubate the eggs (Image 4).

The adjacent wetland had more than 50 Brahminy Kite, Black Kite, and a few feral dogs (Image 5). This place was used as a dumping yard for chicken waste (poultry) (Image 6). Crows were regularly sighted in the vicinity of the WBSE nest, often disturbing and chasing one of the adult WBSE (Image 7). During our observation on 30 January 2023, both adults flew for a few minutes but stayed close to the nests. A few crows sat on the edges of the nest (Image 8) and the WBSEs chased them away. The adult female bird incubated almost throughout the day time. The male often stayed in nearby palm trees and kept a watch on the nest and often chased away nest approaching Brahminy Kites and Black Kites. On 16 February 2023, we observed the presence of two chicks in the nest (Image 9). One was smaller compared to the other chick. The male WBSE brought fish to the nest (Image 10). We also recorded the left-over fish

Image 2. Individual nest width for a comparison with power line structure.

Image 3. Nest construction material.

Image 4. Male White-bellied Sea-Eagle on the nest edge and the female bird incubating in the nest.

Image 5. Brahminy Kite and Black Kite in the nearby wetland used for dumping waste.

Image 6. Poultry waste dumped in the nearest waterbody.

Image 7. Crows chasing White-bellied Sea-Eagle.

Image 8. Crows sitting on the nest edges in the pylon with White-bellied Sea-Eagle.

Image 9. Two chicks of White-bellied Sea-Eagle in the next.

skeletons beneath the nest (Image 11). At times, the fish were taken to the adjacent nest in another nearby pylon and eaten there too. Sometimes, the adult WBSE chased Brahminy Kite and snatched chicken waste from it and brought to the nest for the chicks to feed on. Black Drongo had a good relationship with the WBSE. They were present most of the time on the first and second layers of the pylon and never disturbed the nesting bird.

The breeding season of the WBSEs varies according to location. It occurs in the dry season in Papua New Guinea and from June to August in Australia. According to Ali & Ripley (1974), WBSEs are known to breed from October to January. However, in the Ratnagiri district, nest building occurred from mid-September to January, and chicks were found in the nest by the end of March (Neema et al. 2021). This phenomenon has been documented in more than 70 raptor species worldwide (Hunting 2002; Lehman et al. 2007). Several species of birds are known to use pylons and towers for nesting, perching, and roosting options (Morelli et al. 2014). APLIC (2006) mentions 27 species. Among the bird families, birds of prey are among the groups that are most seriously affected by electrocution (Ellis et al.2009). Habitat destruction represents the most significant threat to the species, as it has resulted in the direct loss of nesting sites and has caused birds to nest in suboptimal habitat types where breeding success can be reduced (Bilney & Emison 1983).

Conclusion

Due to a lack of suitable nesting sites and trees, the WBSE has chosen power line towers for nesting, which are approximately 2 km away from the sea. This helps the bird conveniently scan the marine area for food. It is important to note that the use of man-made structures as nesting sites by the WBSE can pose both risks and benefits to eagles and humans. As a result, careful management and monitoring of these man-made nesting sites are critical to the safety of both eagles and human communities.

Image 10. Male White-bellied Sea-Eagle bringing fish for the chicks.

Image 11. Left over fish remains from beneath the White-bellied Sea-Eagle nesting pylon.

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Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23615–23617

ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)

https://doi.org/10.11609/jott.8453.15.7.23615-23617

#8453 | Received 27 March 2023 | Final received 25 May 2023 | Finally accepted 30 June 2023

NOTE

First record of Horned Grebe *Podiceps auritus* (Linnaeus, 1758) (Aves: Passeriformes: Podicipedidae) from Jammu & Kashmir, India

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The Horned Grebe *Podiceps auritus*, a little diving waterbird in the family Podicipedidae and is found in North America and Eurasia. It breeds in eastern Siberia, western Europe, and Eurasia (Stedman 2020). The bird is a vagrant in India and Pakistan, and migrates during winter (Rasmussen & Anderton 2012; Brraich & Singh 2021). It is reliant on a watery environment throughout the whole year and nests close to the edges of ponds and marshes that have patches of open water and vegetation that emerge from the water. The bird is monogamous and very possessive of its territory. After hatching, the young ones are sub precocial and require care for many days, during which they are fed and kept warm by parents (Stedman 2020). During summer, it primarily feeds on arthropods, including adults and larvae of insects, particularly beetles, dragonflies, mayflies, damselflies, caddisflies, and water bugs. In winter, it mainly forages on fishes and crustaceans except in Europe. In North America, it feeds predominantly on macroinvertebrates (Stedman 2000), while sticklebacks of the family Gasterosteidae are its key prey in Europe (Fjeldså 1973).

The Horned Grebe is widespread in Europe, however, it seems to be relatively uncommon throughout Asia, where it has been considerably less studied; population trends are not known (Stedman 2020). Populations of Horned Grebe are decreasing all over the world (Vlug & Fjeldså 1990), currently it is listed as 'Vulnerable' by the IUCN Red List (Birdlife International 2018). The exact causes of the population decline are unclear, it is quite possible that it is connected to the human disturbance, forestry activities around breeding grounds, competition with sympatric grebes, egg depredation by European Mink *Mustela lutreola*, Raccoon *Procyon lotor*, & Hooded Crow *Corvus cornix*, killing by inclement weather during migration, and loss of natural habitat (del Hoyo et al. 1992; Stedman 2020).

The Wular Lake is an Important Bird Area (IBA) and a well-known Ramsar site in the world. It has a total area of 13,292 ha and is situated about 34 km to the north-west of Srinagar. It is an essential habitat for both migratory as well as resident water birds. The important migratory waterfowl species, including the Common Teal *Anas crecca*, Pintail *Anas acuta*, Shoveler

Editor: H. Byju, Coimbatore, Tamil Nadu, India. **Date of publication:** 26 July 2023 (online & print)

Citation: Zargar, B.N., U. Nazir & Z.H. Najar (2023). First record of Horned Grebe *Podiceps auritus* (Linnaeus, 1758) (Aves: Passeriformes: Podicipedidae) from Jammu & Kashmir, India. *Journal of Threatened Taxa* 15(7): 23615–23617. https://doi.org/10.11609/jott.8453.15.7.23615-23617

Funding: Wildlife Conservation Fund (WCF), Wildlife SOS, and University Grants Commision (UGC).

Competing interests: The authors declare no competing interests.

Acknowledgements: The authors are highly thankful to Wular Conservation and Management Authority (WUCA), Jammu & Kashmir, for granting the necessary permission to carry out birdwatching in Wular lake. A special thanks go out to Mudasir Mehmood Malik (Divsional forest officer Bandipora), Intisar Suhail (Wildlife warden Shopian), and Showket Maqbool, for providing guidance, and support, in identifying this vulnerable bird.

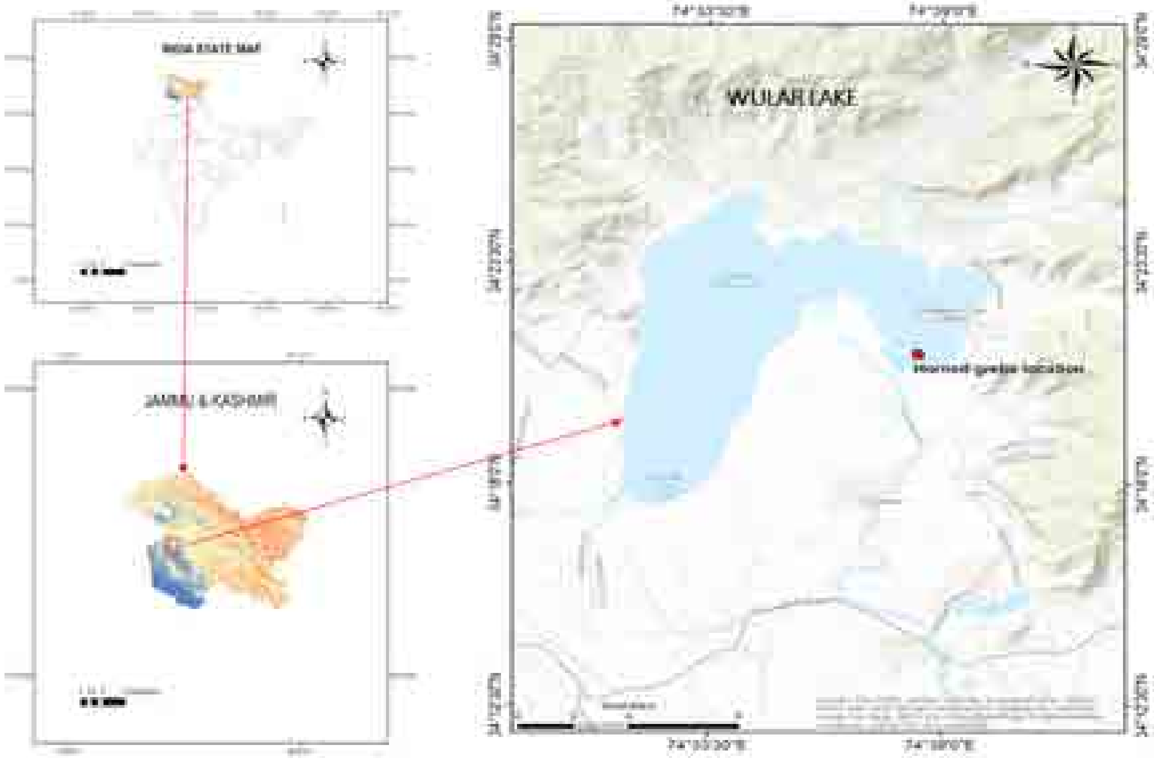

Image 1. The sighting location of Horned Grebe at Wular Lake, Bandipora, Jammu & Kashmir, India.

Anas clypeata, Common Shelduck *Tadorna tadorna*, Ruddy Shelduck *Tadorna ferruginea*, Red-crested Pochard *Netta rufina*, Common Pochard *Aythya ferina*, Mallard *Anas platyrhynchos*, Greylag Geese *Anser anser*, and Long-tailed Duck *Clangula hyemalis*, find the lake to be an ideal wintering location.

The first record of Horned Grebe from India was on 28 December 1993, near Ramnagar, Uttar Pradesh (Drijvers 1995). This was followed by several records from northern India (Brraich & Singh 2021). In India, Horned grebe was mostly seen in Dighal Wetland, Haryana (Ahlawat 2018), and one sighting was also observed in Borit Lake, Hunza Gilgit-Baltistan, Pakistan on 14 December 2016 (Shah 2016; Brraich & Singh 2021). There is no previous published evidence on the presence of Horned Grebe in Jammu & Kashmir including Ladakh (Suhail et al. 2020). Here we report the first record of a group of three Horned Grebes with photographic evidence (Images 2–5). The birds were sighted on 06 March 2023 at 1030 h at Wular Lake of Jammu & Kashmir (Image 1). The birds were feeding and diving during observation. The first two authors successfully got photographs and a short video. The bird was recorded at 34.35327 N, 74.63724 E, with an altitude of 1,597 m. In contrary, the Horned Grebes were seen in December–February in previous sightings observed in other parts of India (Brraich & Singh 2021).

The bird was small in size (31–38 cm) and the bright orange eye suggested (Image 2–5) that it is either a Black-necked or a Horned Grebe. The longish neck, white mark on the lore, head pattern and white tip of the bill differentiate it from the Black-necked Grebe (Mullarney et al. 1999; Prasad 2008). The colour pattern surrounding the eye was also distinctive, the black cap reached only to the centre of the eye, and the line that ran back from the eye was straight. The bill was pale, but the top edge of the upper mandible had a visible black edge, which is a characteristic feature of Horned Grebe.

Regular monitoring of Wular Lake and its surroundings is necessary for wetland conservation and eco-restoration.

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Image 2–5. Horned Grebe *Podiceps auritus* (Linnaeus, 1758) from different angles at Wular Lake, Jammu & Kashmir. © Bilal Nasir Zargar.

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Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23618–23620

OPEN ACCESS

ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)

https://doi.org/10.11609/jott.8512.15.7.23618-23620

#8512 | Received 07 May 2023 | Final received 16 June 2023 | Finally accepted 16 July 2023

NOTE

First photographic record of White Royal *Tajuria illurgis illurgis* (Hewitson, [1869]) (Insecta: Lepidoptera: Lycaenidae) from Arunachal Pradesh, India

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The genus *Tajuria* Moore, [1881] is an Indo-Malayan genus of Blues (Lycaenidae), popularly known as Royal butterflies and comprises of around 50 species across the Oriental tropics (Schröder 2006). India has around 15 species of *Tajuria* so far (Varshney & Smetacek 2015) and out of these, the White Royal *Tajuria illurgis* (Hewitson, 1869), is a rare butterfly (Van Gasse 2021) with no photographic record from Arunachal Pradesh. It was described by Hewitson from Darjeeling, and is also legally protected in India under Schedule II of the Wildlife Protection Act, 1972 (Anonymous 2023). *T. illurgis* (Hewitson, 1869) comprises two subspecies namely, *illurgis*, which has been previously reported from Bhutan, Assam (Basistha et al. 1999), Nepal (Sajan & Sapkota 2022; Van der Poel & Smetacek 2022), northern Thailand (Ek-Amnuay 2012), Laos (Osada et al. 1999), Vietnam (Monastyrskii & Devyatkin 2015), and *tattaka* (Araki, 1949) confined to Taiwan island. Although, Kehimkar (2008) reports its distribution from Uttarakhand to Arunachal Pradesh, Nepal, Bhutan, and Myanmar and it occurs further up to northern Thailand, Laos, Vietnam. Till date, there is no recent photographic evidence of the species from Arunachal Pradesh or any other part of eastern Himalaya in India.

During our field survey in and around Vijaynagar and also inside the forest patches situated behind the small village of Buddhamandir (27.2130^{0}N, 96.9992^{0}E), circle Vijaynagar of Changlang district, Arunachal Pradesh, on 25 August 2022 at 1130h RL photographed a single individual of *Tajuria illurgis illurgis* (Hewitson, [1869]) (Image1) at an elevation of 1,344 m. The butterfly was spotted perching on a leaf of *Strobilanthes* sp. belonging to the family Acanthceae, at a height of about 1.5 m above the ground in shady forest patches, and was observed resting for 7–10 min. No additional observation has been made in the area after repeated survey in subsequent months, which suggest that the species is either rare or highly seasonal at the particular elevation of Vijaynagar. The species is generally known to fly between 1,430–2,200 m across its ranges, and in Nepal it is recently recorded between 1,750–1,860 m (Sajan & Sapkota 2022) and since we recorded the species during August at much lower elevation (1,344 m), it's very likely that the species generally flies lot

Editor: Monsoon Jyothi Gogoi, Ashoka Trust For Research In Ecology And The Environment, Pasighat, India. **Date of publication:** 26 July 2023 (online & print)

Citation: Limbu, R., R. Upadhaya, R. Gogoi & J. Gaur (2023). First photographic record of White Royal *Tajuria illurgis illurgis* (Hewitson, [1869]) (Insecta: Lepidoptera: Lycaenidae) from Arunachal Pradesh, India. *Journal of Threatened Taxa* 15(7): 23618–23620. https://doi.org/10.11609/jott.8512.15.7.23618-23620

Funding: None.

Competing interests: The authors declare no competing interests.

Acknowledgements: The authors expresses sincere gratitude to Mrs. Saranga Limbu, Mrs. Reema Chettri and Mr. Govind Chettri residents of Buddhamandir for helping during the field survey.

Image 1. White Royal *Tajuria illurgis illurgis* (Hewitson, [1869]).

higher elevation during summer at Vijaynagar area. The photograph of the species was identified and confirmed following (Kehimkar 2008; Ek-Amnuay 2012).

The previous detailed studies on the taxonomy of butterflies from Arunachal Pradesh (Bhattachacharya 1985; Gogoi 2012; Sethy et al. 2014; Durairaj & Sinha 2015; Singh 2015; Kehimkar 2016; Sondhi & Kunte 2016; Sharma & Goswami 2021) revealed no published records of *Tajuria illurgis illurgis* (Hewitson, [1869]) from the state of Arunachal Pradesh. Therefore, this is the first photographic evidence of *Tajuria illurgis illurgis* (Hewitson, [1869]), White Royal from Arunachal Pradesh. The presence of *T. illurgis* in Arunachal Pradesh was not unexpected as it is historically known to occur all throughout Himalayan ranges from Uttarakhand-Bhutan-Myanmar (Van Gasse 2021). The encounter of this species which has received India's highest level of legal protection, Vijaynagar shows that more scientific exploration needs to be carried out from the area, and being in the extreme boundary of Indo-Myanmar, newer Lepidopteran findings can come up in future.

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Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23621–23626
ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)
https://doi.org/10.11609/jott.8344.15.7.23621-23626
#8344 | Received 28 December 2022 | Final received 19 May 2023 | Finally accepted 31 May 2023

NOTE

Preliminary observations of moth fauna of Purna Wildlife Sanctuary, Gujarat, India

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Gujarat is the fifth largest state of India and is situated on the western coast with a coastline of 1,600 km under the Kathiawar peninsula. There are 33 districts in Gujarat. Purna Wildlife Sanctuary (Dang District, Gujarat), known as a hotspot for its biodiversity, is situated on the extreme northern side of the Western Ghats. It has tropical moist deciduous forests with various flora and fauna in it. It comprises two protected areas — Purna Wildlife Sanctuary (WS) and Vansda National Park. They are known to protect the precious fauna of the area but limited information is available on the invertebrate fauna from the sanctuary. Purna WS is rich in its fauna because of its different terrain, landscapes, and forest.

Purna Wildlife Sanctuary is located at Dang District of Gujarat under the coordinates 20.91793°N, 73.7007°E with an area of 160.84 km^2. It has southern moist deciduous forests and southern dry deciduous forests (Champion & Seth 1968; Singh et al. 2000), with a normal rainfall of 1,600 mm annually. The topography of the WS is undulant with an altitude range of 130–1100 m. Thus, the WS has a varied range of flora and fauna. Moths play an important role as an indicator of the environmental health of an ecosystem (Bachanda et al. 2014). Moth larvae are herbivores, pests of vegetables, and crops, thus playing ecological roles throughout the life cycle (Scriber & Feeny 1979) while adults and larval stages are food sources for other animals and some are night pollinators (Holt 2002; Hahn & Bruhl 2016).

In class Insecta, moths are among the most varied groups (Soggard 2009). There are almost 1,65,000 species of moths throughout the world (Khan 2018), out of which about 12,000 species are described from India (Chandra & Nema 2007; Bell & Scott 1937; Cotes & Swinhoe 1887–1889; Hampson 1892, 1894, 1895, 1896; Chandra 2007; Gurule & Nikam 2013; Smetacek 2011; Uniyal et al. 2013; Sondhi & Sondhi 2016). Four-hundred-and-one species of moths have been recorded from Gujarat (Nurse 1899; Mosse 1929; Gupta & Thakur 1990). Further, no information is available on the moths from the Purna WS and therefore the present study was conducted for the first time.

The survey of Purna WS was carried out from 2019 to 2022. Various localities were visited—Bardipada range, Bheskatri range, Kalibel range, and Singhana range of Dang & Ahwa districts of Gujarat (Table 1)—and for the collection, night traps for about 5–6 hours was used for trapping moths at night.

Observation and collection of moths was done using a mercury vapour bulb of 200W on a white sheet. A collection permit for moths was received from the Gujarat Forest Department vide letter no. WLP/RES/28/C/119-120/2020-21 dated 01/09/2020.

Editor: Sanjay Sondhi, Titli Trust, Dehradun, India. **Date of publication:** 26 July 2023 (online & print)

Citation: Choudhary, P. & I. Sharma (2023). Preliminary observations of moth fauna of Purna Wildlife Sanctuary, Gujarat, India. *Journal of Threatened Taxa* 15(7): 23621–23626. https://doi.org/10.11609/jott.8344.15.7.23621-23626

Funding: Ministry of Environment, Forest & Climate Change (MoEF & CC), Zoological Survey of India.

Competing interests: The authors declare no competing interests.

Acknowledgements: Authors are thankful to the director Dr. Dhriti Banerjee, Zoological Survey of India for providing the necessary facilities and encouragement.

Figure 1. The surveyed area of Dangs District, Gujarat.

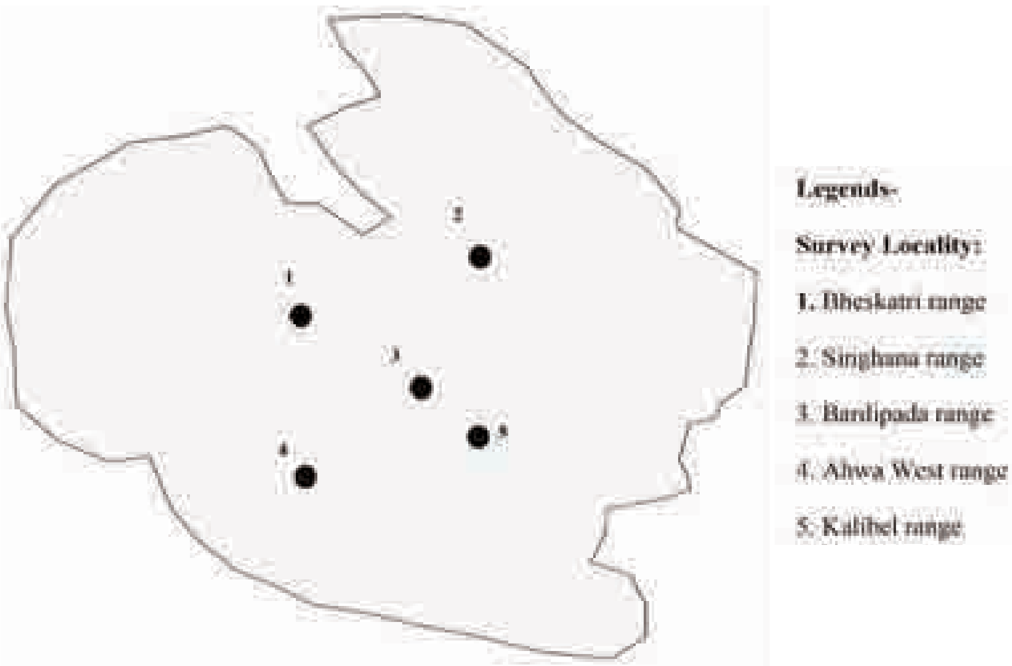

Figure 2. The survey localities of Purna Wildlife Sanctuary.

Collected specimens were labeled with locality labels in the field. Later on, they were sorted, relaxed, pinned, identified up to the species level, and labelled. Their identification was done with the help of identification keys, standard reference books, and available literature. Further specimens are deposited at the National Zoological Collection of Desert Regional Centre, Jodhpur. Four-hundred-and-seven moth specimens were

Table 1. Collection of data from various localities of the study area.

	District	Sites surveyed	Exs. collected
1.	Dang	Bardipada range	153
2.		Bheskatri range	26
3.		Kalibel range	141
4.		Singhana range	48
5.	Ahwa	Ahwa West range	39
		Total	**407**

Table 2. List of preliminary observation moth fauna from Purna Wildlife Sanctuary.

	Scientific name	Status
Super family: Pyraloidea **Family:** Crambidae		
1	*Botyodes asialis* Guenée , 1854	Common
2	*Conogethes punctiferalis* (Guenée , 1854)	Rare
3	*Cydalima laticostalis* (Guenée , 1854)	Common
4	*Diaphania indica* (Saunders, 1851)	Common
5	*Parotis marginata* (Hampson, 1893)	Rare
Super family: Noctuoidea **Family:** Erebidae		
6	*Achaea janata* (Linnaeus, 1758)	Common
7	*Amata cyssea* (Stoll, [1782])	Rare
8	*Anomis flava* Fabricius, 1775	Rare
9	*Argina astrea* (Drury, 1773)	Common
10	*Arna bipunctapex* (Hampson, 1891)	Rare
11	*Asota caricae* (Fabricius, 1775)	Common
12	*Asota ficus* (Fabricius, 1775)	Common
13	*Chalciope mygdon* (Cramer, [1777])	Common
14	*Creatonotos gangis* (Linnaeus, 1763)	Common
15	*Eudocima phalonia* (Linnaeus, 1763)	Common
16	*Lymantria serva* (Fabricius, 1793)	Rare
17	*Lyncestis amphix* (Cramer, 1777)	Rare
18	*Nepita conferta* (Walker, 1854)	Rare
19	*Orvasca subnotata* Walker, 1865	Rare
20	*Perina nuda* (Fabricius, 1787)	Common
21	*Spilarctia* sp.	Rare
22	*Spirama helicina* (Hubner, 1824)	Common
23	*Sphrageidus similis* (Fuessly, 1775)	Common
24	*Syntomoides imaon* (Cramer, [1779])	Common
25	*Thyas coronata* Fabricius (1775)	Common
26	*Thyas honesta* Hübner, [1824]	Common
27	*Trigonodes disjuncta* (Moore, 1882)	Common
28	*Utetheisa lotrix* (Cramer, 1779)	Common
Family: Noctuidae		
29	*Spodoptera litura* (Fabricius, 1775)	Common
Super family: Geometroidea **Family:** Geometridae		
30	*Biston suppressaria* (Guenée, [1858])	Rare
31	*Hypomecis* sp.	Rare
Super family: Lasiocampoidea **Family:** Lasiocampidae		
32	*Trabala ganesha* Roepke, 1951	Rare
33	*Trabala vishnou* Lefebvre, 1827	Rare
Super family: Pyraloidea **Family:** Pyralidae		
34	*Cadra cautella* (Walker, 1863)	Rare
Super family: Bombycoidea **Family:** Saturniidae		
35	*Actias selene* (Hübner, [1807])	Rare
36	*Antheraea paphia* (Linnaeus, 1758)	Rare
Super family: Bombycoidea **Family:** Sphingidae		
37	*Daphnis nerii* (Linnaeus, 1758)	Common
38	*Marumba dyras* (Walker, 1856)	Common
39	*Nephele hespera* (Fabricius, 1775)	Common
40	*Psilogramma* sp.	Common
41	*Theretra nessus* (Drury, 1773)	Rare
Super family: Zygaenoidea **Family:** Limacodidae		
42	*Parasa lepida* (Cramer, 1799)	Rare

collected and identified to 42 species under 39 genera and nine families. During the study, it was found that Erebidae is a dominant family of moths followed by Sphingidae, Crambidae, Saturniidae, Geometridae, Lasiocampidae, Noctuidae, Limacodidae, and Pyralidae in Purna Wildlife Sanctuary.

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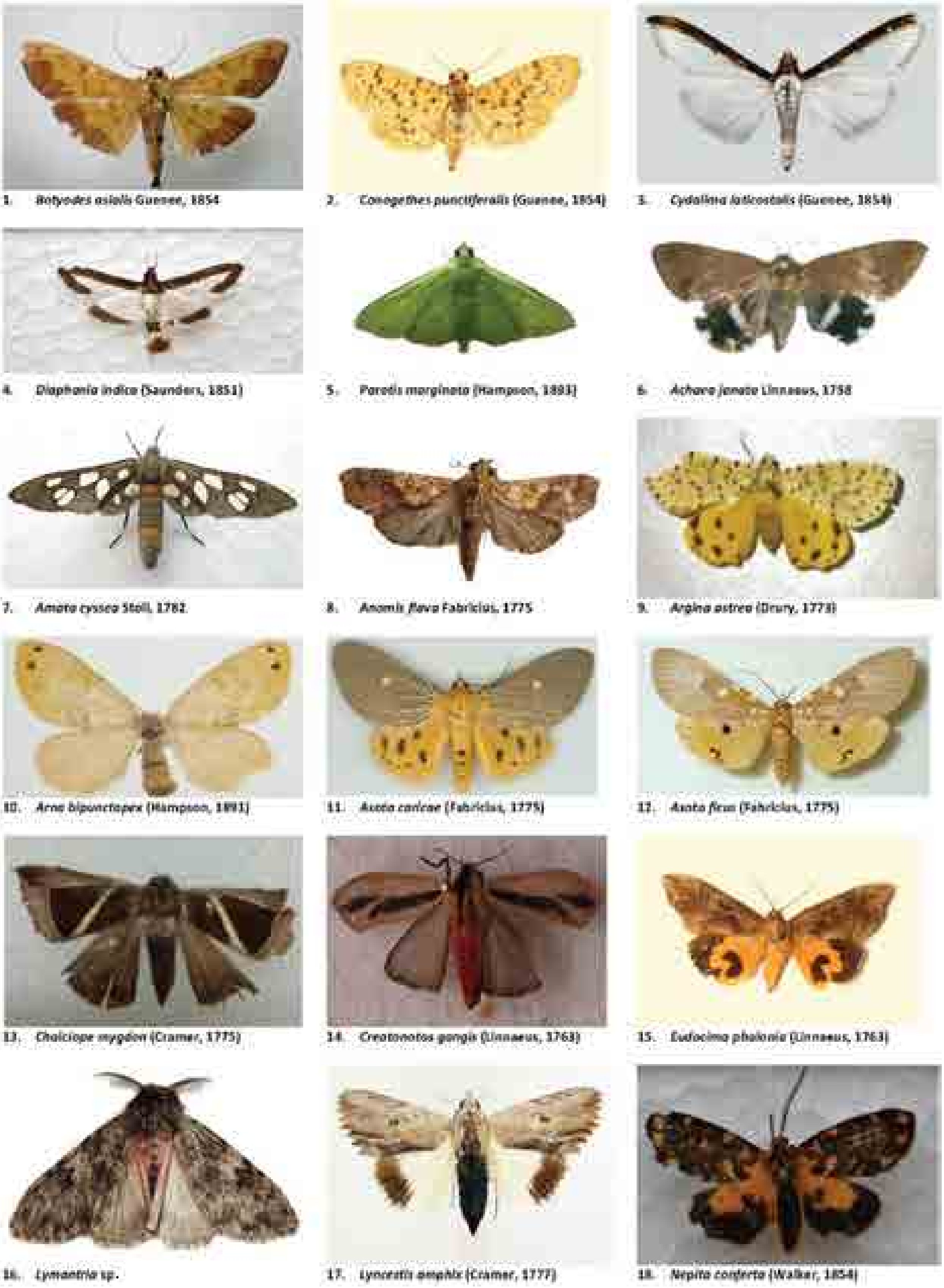

1. *Botyodes asialis* Guenee, 1854
2. *Conogethes punctiferalis* (Guenee, 1854)
3. *Cydalima laticostalis* (Guenee, 1854)
4. *Diaphania indica* (Saunders, 1851)
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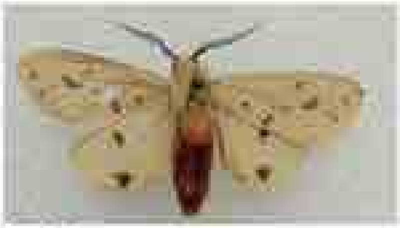
14. *Creatonotos gangis* (Linnaeus, 1763)
15. *Eudocima phalonia* (Linnaeus, 1763)
16. *Lymantria* sp.
17. *Lyncestis amphix* (Cramer, 1777)
18. *Nepita conferta* (Walker, 1854)

Image 1–18. Some moths of Purna Wildlife Sanctuary.

19. *Orvasca subnotata* Walker, 1865

20. *Perina nuda* Fabricius, 1787



21. *Spilosoma* sp.

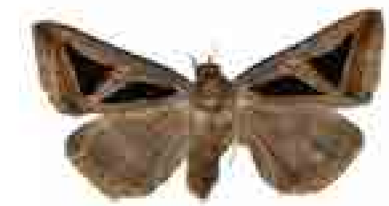
22. *Spirama helicina* (Hubner, 1824)

23. *Sphrageidus similis* (Fussli, 1775)

24. *Syntomoides imaon* (Cramer, 1775)

25. *Thyas coronata* Fabricius (1775)

26. *Thyas honesta* Hubner, 1824



27. *Trigonodes disjuncta* (Moore, 1882)

28. *Utetheisa lotrix* Cramer, 1779

29. *Biston suppressaria* (Guenee, 1857)

30. *Hypomecis* sp.

31. *Trabala ganesha* Roepke, 1951

32. *Trabala vishnou* Lefebvre, 1827

33. *Cadra cautella* (Walker, 1863)

34. *Actias selene* (Hübner, 1807)

35. *Antheraea paphia* Linnaeus, 1758

36. *Daphnis nerii* (Linnaeus,1758)

Image 19–36. Some moths of Purna Wildlife Sanctuary.

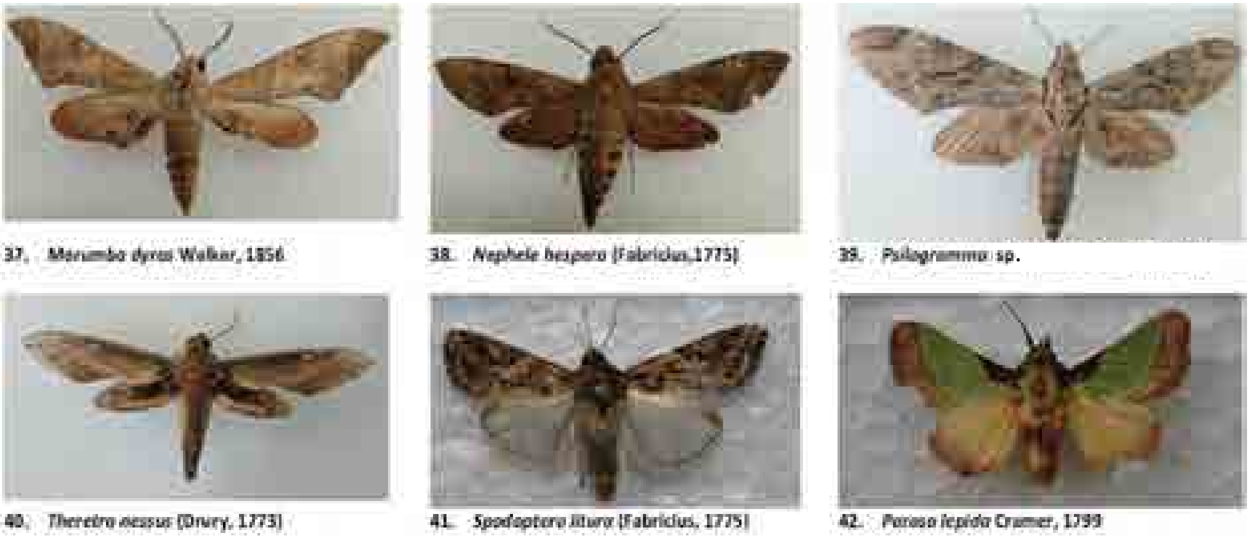


Image 37–42. Some moths of Purna Wildlife Sanctuary.

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Journal of Threatened Taxa | www.threatenedtaxa.org | 26 July 2023 | 15(7): 23627–23630
ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)
https://doi.org/10.11609/jott.8196.15.7.23627-23630
#8196 | Received 16 September 2022 | Final received 15 June 2023 | Finally accepted 28 June 2023

NOTE

Argyreia lawii C.B.Clarke (Convolvulaceae) – an extended distribution record in the Western Ghats of Kerala

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Argyreia Lour. is considered to be one of the largest and complex genus among the family Convolvulaceae. It consists of around 135 taxa (Staples & Traiperm 2017) distributed in southeastern Asia, China, and in the Indian subcontinent. In India, the genus is represented with 40 species and considered to be the second most species abundant genus among Convolvulaceae (Lawand et al. 2019).

During the floristic survey in Attappady area at Palakkad District, Kerala, collected an interesting species of *Argyreia* at a specific location along the way of Thavalam, an area 18 km away from Silent Valley which comes under wet evergreen forest. The specimen was collected with flowers and the identity was confirmed as *A. lawii* by Botanical Survey of India (BSI), Southern Regional Centre (SRC), Coimbatore. The sample specimen was stored in Avinashilingam Institute Herbarium, for further use. While checking for the distribution of the species it is previously known only from Karnataka (Gamble 1922), recently rediscovered from Maharashtra (Lawand et al. 2019) and Shalini et al. (2018) added to the flora of Tamil Nadu. Other than this it is not reported anywhere else in India including Kerala (Kumar et al. 2005; Nayar et al. 2014; Eflorakerala). Hence the present collection from Palakkad District of Kerala shows the extended distribution of the species and addition to the state flora as well. A short description along with color photographs are provided here to facilitate the future identification and collection (Images 1 & 2).

Argyreia lawii C.B.Clarke in Hook.f., Fl. Brit. India 4: 190. 1883; T.Cooke, Fl. Bombay 2: 327. 1908; Gamble, Fl. Pres. Madras 2: 908. 1922; B.D.Sharma et al., Fl. Karnataka 179. 1984; M.R.Almeida, Fl. Maharashtra 3: 310. 2001; Venakanna & Das in N.P.Singh et al., Fl. Maharashtra 2: 445. 2001; Shalini et al., Indian J. Forest. 41(3): 265–268. 2018.

Description

A semi-woody climber, the stem is strigose, terete, greenish, and herbaceous. Leaves simple, alternate, and elliptic-ovate, 6–10.2 x 3–5.5 cm, base rounded, acute apex, and entire margin. Strigose on both the surface, midrib conspicuous with lateral veins 7–8 pairs. Petiole is about 1.7–3.8 cm, cylindrical, strigose, stout, and wooly. Inflorescence is an axillary cyme compacted with 5–7 flowers, dichotomously branched with one central flower. Peduncle 3–6 cm long, longer than petiole, terete, and less strigose. Flowers sub-sessile, bracteate, whorls slightly strigose, inner whorl narrows than the

Editor: Anonymity requested. **Date of publication:** 26 July 2023 (online & print)

Citation: Rajeswari A.R. & M.K. Nisha (2023). *Argyreia lawii* C.B.Clarke (Convolvulaceae) – an extended distribution record in the Western Ghats of Kerala. *Journal of Threatened Taxa* 15(7): 23627–23630. https://doi.org/10.11609/jott.8196.15.7.23627-23630

Funding: None.

Competing interests: The authors declare no competing interests.

Acknowledgements: The authors are thankful to the head of office, Botanical Survey of India, Southern Regional Centre, Coimbatore for permitting us to consult the herbaria and library.

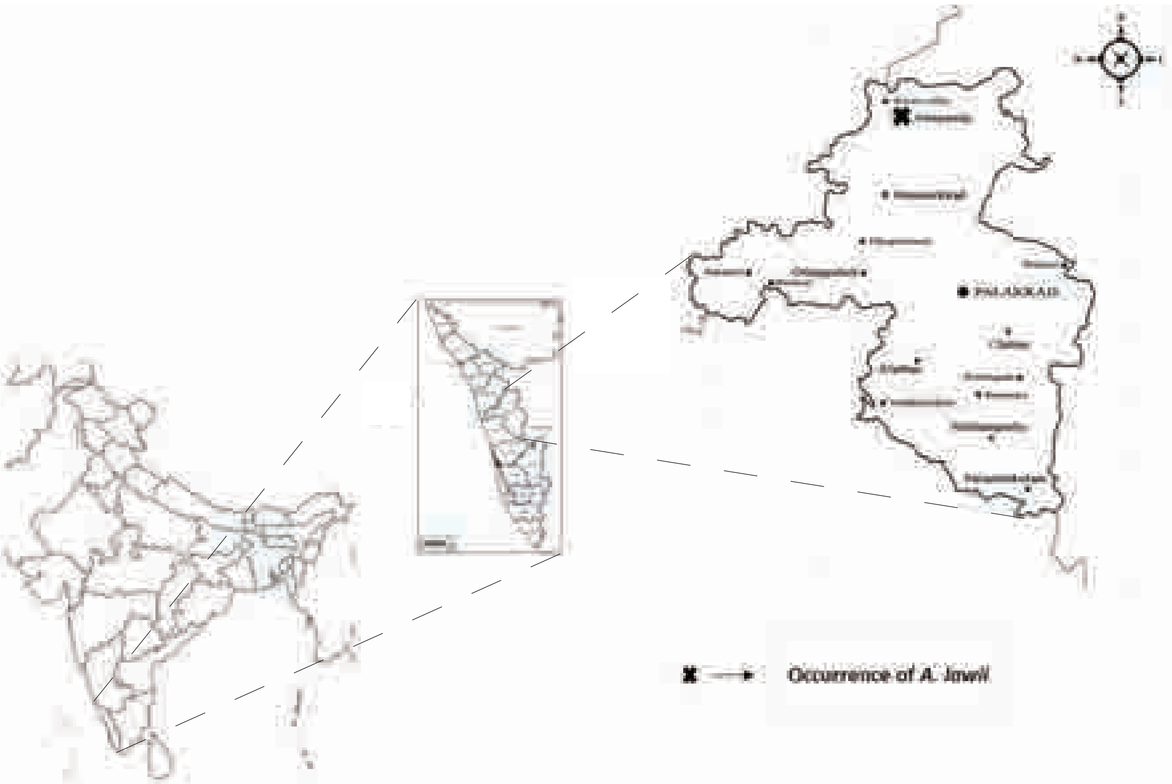

Figure 1. Distribution of *Argyreia lawii* C.B.Clarke in Attappady, Palakkad District, Kerala, India.

Image 1. *Argyreia lawii*: A—Habitat | B—Habit closeup | C—Inflorescence | D—Twig | E—Corolla front view. © A.Raja Rajeswari.

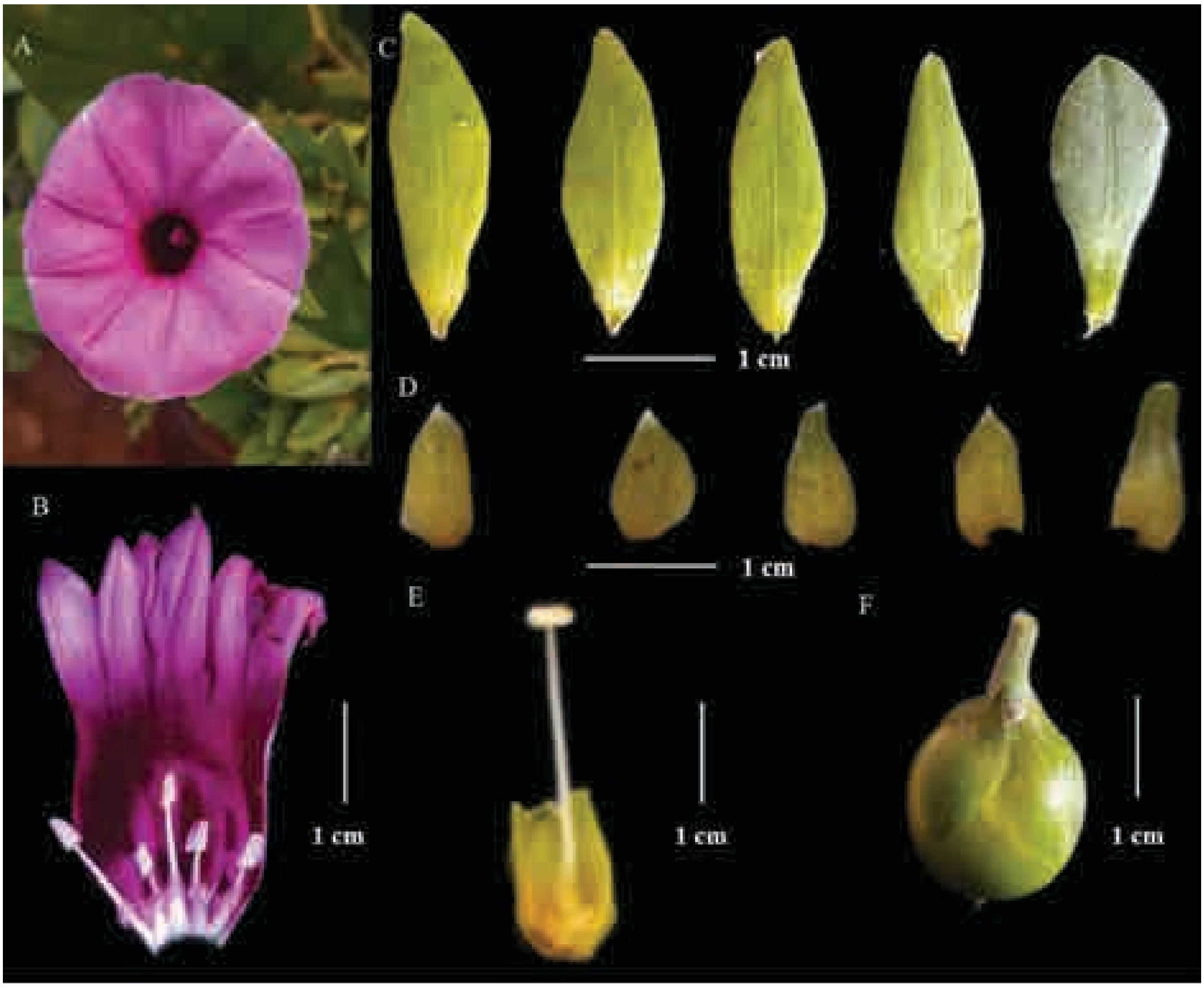

Image 2. *Argyreia lawii*: A—Corolla closeup | B—opened flower | C—Bracts | D—Sepals | E—Gynoecium | F—Fruit. © A.Raja Rajeswari.

outer and oblong. Bracts oblong or elliptical 1–2.7 cm x 0.4–0.8 cm, strigose, outer bracts are wider than the inner ones. Sepals 5, subequal shorter than bracts 8–10 x 5–6 mm, ovate, and acute apex, glabrous to pubescent, gamosepalous. Corolla infundibulum 3.5–4.7 cm x 1.8–2.8, hairy, disc slightly 5-lobed. Stamens 5; filaments 5, unequal, 2 long 1.6–1.8 cm, 3 short 1–1.2 cm, adnate, above the base of the corolla. Ovary conical, glabrous, style, separately dilated, jointed at base longer than filament 1.4–1.9 cm or sometimes unequal. Stigma papillated and bilobed. Fruit is a berry with 5 persistent calyx lobes, young green, when matured yellow.

Flowering and Fruiting: May to August.

Habitat and ecology: Twining shrub along roadside margins of wet evergreen forest at an elevation of 662 m growing in association with species like *Asystasia gangetica* (L.) T.Anderson, *Cardiospermum halicacabum* L., *Causonis trifolia* (L.) Mabb. & J.Wen, *Chromolaena odorata* (L.) R.M.King & H.Rob., *Justicia adhatoda* L., *Lantana camara* L., *Mesosphaerum suaveolens* (L.) Kuntze, *Mimosa pudica* L., *Oplismenus compositus* (L.) P.Beauv., *Parthenium hysterophorus* L., and *Rotheca serrata* (L.) Steane & Mabb. We could observe 12–15 mature individuals covering the total area that may not exceed 5 km^2.

Distribution: Karnataka (Western Ghats region, Konkan Province & Bababudhan Hills of Karnataka State), Kerala (Present report – Thavalam, Palakkad District), Maharashtra (Bhudargad Fort & Patgaon, Kolhapur District), and Tamil Nadu (Nilgiris District, Coonoor Ghat).

Specimen examined: India, Kerala, Palakkad District; Thavalam, 13.120^{0}N, 76.591^{0}E, 22.08.2022, A. Raja Rajeswari ARR0001, Avinashilingam Institute Herbarium (Image 3).

Notes: *Argyreia lawii* C.B.Clarke may be facing

Image 3. Herbarium of *Argyreia lawii* [#A Raja Rajeswari ARR0001(TAK014)].

threats due to the widening of road, domination of exotic plants, removal of plants along the road side and cultivated fields by the local community in Thavalam area.

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NAAS rating (India) 5.64

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OPEN ACCESS

ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)

July 2023 | Vol. 15 | No. 7 | Pages: 23463–23630
Date of Publication: 26 July 2023 (Online & Print)
DOI: 10.11609/jott.2023.15.7.23463-23630

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