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# Documentation and Comparison of Amphibian Diversity in Natural Montane Forest Ecosystem and Human-Modified Tea Garden Ecosystem in the Darjeeling Himalayas, West Bengal, India: Implications for Habitat Conservation

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## ABSTRACT

This study documents and compares amphibian diversity in two distinct ecosystems in the Darjeeling Himalayas: a montane forest ecosystem (Chattakpur forest) and an adjacent human-modified tea garden agroecosystem (Ringtong Tea Estate), using standardized visual encounter surveys conducted from May to October 2025. Eleven amphibian species (two orders, six families) were recorded across both sites. Species composition, richness, and diversity indices were analyzed to quantify differences. Each ecosystem supported nine species, but the tea garden had a higher total abundance (181 individuals) than the forest (148). Beta diversity was calculated to analyze compositional turnover between sites. Photographic documentation and conservation assessments confirmed the presence of several near-threatened species in both habitats. These findings challenge the notion that anthropogenic landscapes invariably reduce amphibian diversity and underscore the conservation value of shade-grown tea plantations in the Eastern Himalayan biodiversity hotspot.

**Keywords** Amphibian diversity, Eastern Himalayas, Tea garden, Beta diversity, Conservation, Montane forest, Alpha diversity, Anthropogenic disturbances, Biodiversity hotspot.

## INTRODUCTION

Amphibians are poikilothermic tetrapods (Girard *et al.* 2015) with biphasic life cycles that link aquatic and terrestrial ecosystems (Rozo and Camacho-Reyes 2022), making them sensitive bioindicators of environmental health (Amado and Megia-Palma 2024). The biphasic life cycle of most amphibians underscores their value in studying wetland functionality and connectivity (Jeliazkov *et al.* 2019; Zamberletti *et al.* 2018). As active predators of invertebrates, they regulate insect populations (Syazali *et al.* 2019), while serving as prey, they support higher trophic levels (Zipkin and DiRenzo 2022). Despite decades of study, many amphibian species remain undescribed, particularly in biodiversity hotspots (Carné and Vie-

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ites 2024). Globally, 8,649 amphibian species are currently recognized, with anurans dominating taxonomic diversity (Frost 2026). In India, the Zoological Survey of India has documented 454 species as of 2023 Dinesh *et al.* (2023), and West Bengal accounts for 49 of them (Deuti 2021). Within West Bengal, the Darjeeling Himalayan region plays a pivotal role in amphibian conservation, harboring 37 species across 18 genera, 8 families, and 3 orders (De 2016). This richness is driven by pronounced altitudinal gradients from subtropical lowlands to alpine zones, creating heterogeneous microhabitats supported by high annual precipitation, which are ideal for both breeding and non-breeding phases (Pradhan *et al.* 2018).

The Darjeeling region lies within the Eastern Himalayas, one of the world's biodiversity hotspots (Chettri 2010). Despite its documented amphibian diversity, the area remains understudied relative to adjacent regions such as Sikkim and Nepal (Rana *et al.* 2021). Recent taxonomic advancements, including integrative studies combining morphology, bioacoustics, and molecular markers (Mandal *et al.* 2025; Boruah *et al.* 2025), emphasise the urgency of continued surveys to uncover hidden diversity and resolve complex species relationships.

This study addresses a critical knowledge gap by comparing amphibian diversity between a natural montane forest ecosystem (Chattakpur Forest) and an adjacent human-modified tea garden ecosystem (Ringtong Tea Estate) in the Darjeeling Himalayan Region. By quantifying species composition, relative abundances, alpha and beta diversity, and habitat associations, the research evaluates the influence of anthropogenic factors on amphibian assemblages—the strategies in the eastern Himalayan biodiversity hotspot, where tea plantations occupy large areas. Ultimately, the study tests whether a responsibly managed tea garden ecosystem can serve as a complementary refuge for declining amphibian populations amid climate change and land-use pressures.

## MATERIALS AND METHODS

### Study area

Surveys were conducted in Chattakpur Forest (26.937308°N, 88.311198°E, approximately 2161 m) and Ringtong Tea Estate (26.923478°N, 88.272652°E, approximately 1537m), Kurseong subdivision, Darjeeling district (Fig. 1). Both sites receive >2500mm annual rainfall, predominantly during the monsoon (June-September). The Chattakpur forest features pine-dominated vegetation with seasonal streams, while the tea garden contains *Camellia sinensis* monoculture interspersed with spade trees, bamboo and rain-fed puddles. The Ringtong Tea Estate's Rainforest Alliance certification implies that specific agricultural practices, such as maintaining canopy cover and using reduced chemical inputs (semi-organic), are in place.

### Data collection

Fieldwork was carried out from May to October 2025 in the Ringtong tea garden and the Chattakpur forest (30 visits in total), with equal sampling effort at both sites. Amphibians were surveyed using a time-constrained visual encounter method (Heyer *et al.* 1994) and acoustic encounter surveys. Most sampling was conducted during daylight hours and in the evenings, since amphibians are nocturnal (4-5 hours per visit). The surveys involved inspecting various habitats, such as under boulders, leaves, bushes, and vegetation, as well as rain-fed puddles and nearby streams, to detect amphibians. Individuals of the same species observed multiple times were not recounted to avoid pseudo-replication. All observations were photographed for identification, and no specimens were collected. Species were identified using keys from Dinesh *et al.* (2023), De (2016), and Mandal et al. (2025). Surveys were conducted in accordance with ethical guidelines for non-invasive amphibian research.

### Data analysis

All the amphibian data were first compiled at the site level, including species presence-absence and abundance. Within each site, alpha diversity was calculated in PAST v4.15 to assess species richness, evenness, and heterogeneity using the following indices: Shannon diversity index (H′) to account for both richness and evenness ($H' = -\Sigma$ pi ln pi), evenness ($J=H'/\ln S$), dominance, and Simpson's index (1-D).

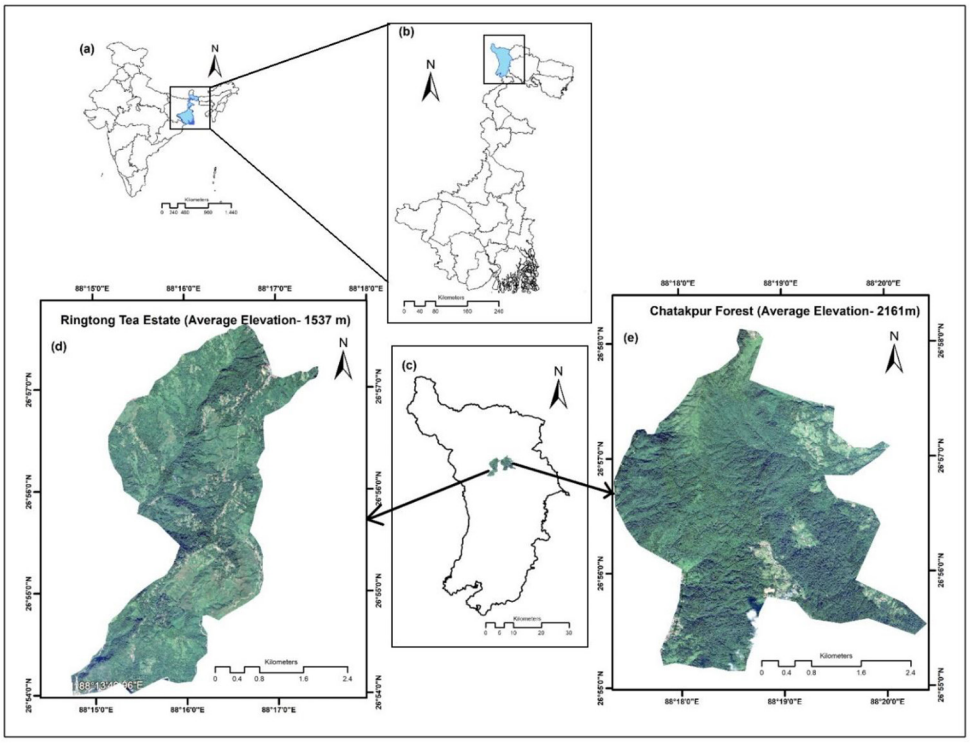



**Fig. 1.** Map showing the position of the study area **(a).** West Bengal in relation to India, **(b).** position of District Darjeeling in respect to West Bengal, **(c).** Location of the study sites within District Darjeeling, **(d).** Ringtong Tea Estate with average elevation, **(e).** Chattakpur Forest with average elevation. (ArcGIS 10.4.1 was used to create this map).

**Table 1.** Absolute and relative abundance of amphibian species with their conservation status.

| Species recorded | Chattakpur forest (n) | Relative abundance in forest (%) | Ringtong tea garden (n) | Relative abundance in tea garden (%) | Total individuals | Overall % | IUCN status | Wildlife (Protection) Act schedule in India (2022) |
|---|---|---|---|---|---|---|---|---|
| *Duttaphrynus himalayanus* | 25 | 16.89 | 31 | 17.13 | 56 | 17.02 | LC | Not listed |
| *Nanorana liebigii* | 10 | 6.76 | 13 | 7.18 | 23 | 6.99 | LC | Schedule II |
| *Nanorana annandalii* | 23 | 15.54 | 25 | 13.81 | 48 | 14.59 | NT | Not listed |
| *Euphlyctis cyanophlyctis* | 0 | 0.00 | 2 | 1.10 | 2 | 0.61 | LC | Schedule II |
| *Megophrys robusta* | 5 | 3.38 | 2 | 1.10 | 7 | 2.13 | LC | Not listed |
| *Megophrys major* | 2 | 1.35 | 0 | 0.00 | 2 | 0.61 | LC | Schedule II |
| *Amolops marmoratus* | 20 | 13.51 | 35 | 19.34 | 55 | 16.72 | LC | Not listed |
| *Raorchestes annandalii* | 43 | 29.05 | 23 | 12.71 | 66 | 20.06 | LC | Not listed |
| *Polypedates maculatus* | 0 | 0 | 16 | 8.84 | 16 | 4.86 | LC | Not listed |
| *Polypedates himalayanus* | 13 | 8.78 | 0 | 0.00 | 13 | 3.95 | LC | Not listed |
| *Tylototriton himalayanus* | 7 | 4.73 | 34 | 18.78 | 41 | 12.46 | NT | Schedule I |

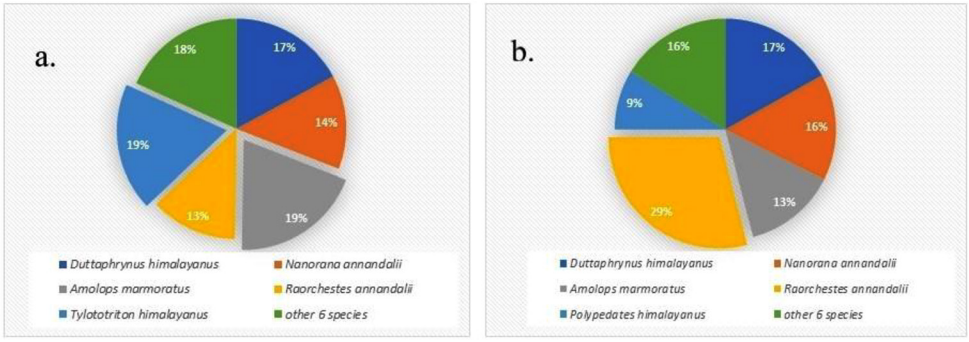



**Fig. 2.** Comparison of relative frequency between forest and tea garden ecosystems: **(a).** Chattakpur forest ecosystem, **(b).** Ringtong tea garden ecosystem.

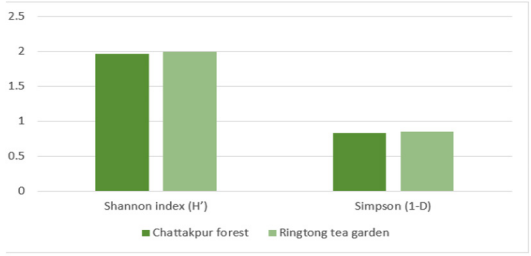



**Fig. 3.** Comparison of Shannon index (H') and Simpson index (1-D) between two ecosystems.

Beta diversity was calculated using the Jaccard and Sorensen indices (J = a/(a+b+c), Sorensen = 2a/(2a+b+c). Bray-Curtis dissimilarity was computed on the presence-absence matrix for comparability with Jaccard/ Sorensen. Gamma diversity was the total unique species across both sites.

**Species composition and abundance:** Eleven species were recorded (Table 1). Both sites supported nine species each. The forest recorded 148 amphibian




**Fig. 4.** Photographic documentation of the amphibians recorded during the study.
**(a).** Duttaphrynus himalayanus, **(b).** Raorchestes annandalii, **(c).** Polypedates himalayanus, **(d).** Megophrys robusta, **(e).** Nanorana liebigii, **(f).** Megophrys major, **(g).** Polypedates maculatus, **(h).** Nanorana annandalii, **(i).** Euphlyctis cyanophlyctis, **(j).** Amolops marmoratus, **(k).** Tylototriton himalayanus.

individuals, whereas the tea garden recorded 181. Dominant species differed: *Raorchestes annandalii* (29.05%) in the forest, whereas *Amolops marmoratus* (19.34%) and *Tylototriton himalayanus* (18.78%) in the tea garden (Fig. 2).

**Alpha Diversity:** Species richness was identical (9 per site), but abundance and evenness were higher in the tea garden (H′=1.995, 1-D=0.8543) than in the forest (H′= 1.959, 1-D=0.8344). (Fig. 3).

**Beta and Gamma Diversity:** Beta diversity indicated moderate similarity (Jaccard=0.636; Sorensen=0.778; Bray-Curtis dissimilarity=0.669). Gamma diversity = 11 species (7 shared; 2unique per site).

**Photographic Documentation and Conservation Status**

All 11 species were photographed in situ (Fig. 4). Conservation assessments (IUCN) listed *Tylototriton himalayanus* as Vulnerable Schedule I. Several anurans are Schedule II or Data Deficient (Table 1).

## DISCUSSION

The present study provides the first quantitative comparison of amphibian diversity between a natural montane forest ecosystem and an adjacent human-modified tea garden ecosystem in the Darjeeling Himalayas. Despite identical species richness (9 species per site), the tea garden supported higher total abundance and marginally higher alpha diversity (Shannon-Wiener H′=1.995 and Simpson's 1-D = 0.8543 in the tea garden and H′=1.959 and 1-D = 0.8344 in the forest). Beta diversity indices indicated moderate compositional turnover (Jaccard similarity = 0.636; Sorensen similarity = 0.778; Bray-Curtis dissimilarity = 0.669), with seven species shared between sites and two unique to each habitat. This pattern contrasts with the widespread global trend of amphibian decline in the human-modified landscapes (Landler *et al.* 2023).

Several site-specific factors appear to drive this elevated abundance. The hydrological regime in Ringtong tea estate is characterized by an extensive network of irrigation channels and numerous rain-fed puddles that retain water well into the post-monsoon period. These lentic micro-waterbodies are ideal breeding sites for generalised anurans such as *Euphlyctis cyanophlyctis* and *Polypedates maculatus* whose larvae thrive in still or slow-moving water (Murali and Raman, 2012) with abundant microorganisms and algae (Mogali *et al.* 2022). In contrast, Chattakpur forest is dominated by fast-flowing hill streams that may experience flash flooding, which can reduce larval survival for many species due to increased drift and mortality (Kiss *et al.* 2022; Walls *et al.* 2013).

Dense natural forests with greater tree diversity can harbor more diverse predators, leading to stronger top-down control (Stemmelen *et al.* 2021; Vázquez-González *et al.* 2024). In human-modified forest systems and agro-forest ecosystems, maintaining a complex local vegetation structure—such as a higher density of understory shrubs like the tea bushes, fallen trunks, and a dense canopy from shade trees—stabilises microclimatic conditions, broadens available ecological niches, and provides key microhabitats that foster greater herpetofauna diversity (Cervantes-López *et al.* 2025).

Although both sites are located within the same altitudinal belt and experience similar climatic regimes (>2,500 mm annual rainfall), the nature and intensity of human disturbance differ markedly. In Chattakpur forest, anthropogenic pressures include garbage dumping, religious idol immersion in seasonal streams, and occasional subsistence hunting, factors known to degrade water quality and directly reduce amphibian populations (Pradhan *et al.* 2018; Chettri *et al.* 2020).

In the Ringtong Tea Estate, management practices appear relatively benign; pesticide application is limited, shade trees are retained, and streams/puddles remain largely free of solid waste. The resulting hydrological stability favors generalist pond-breeding species, notably the Himalayan newt (*Tylototriton himalayanus*) and *Amolops marmoratus*. Moreover, adjacent streams within the forest receive direct influxes of garbage from nearby human settlements (Fig. 5a). This pattern aligns with the "matrix quality" hypothesis in landscape ecology, which posits that

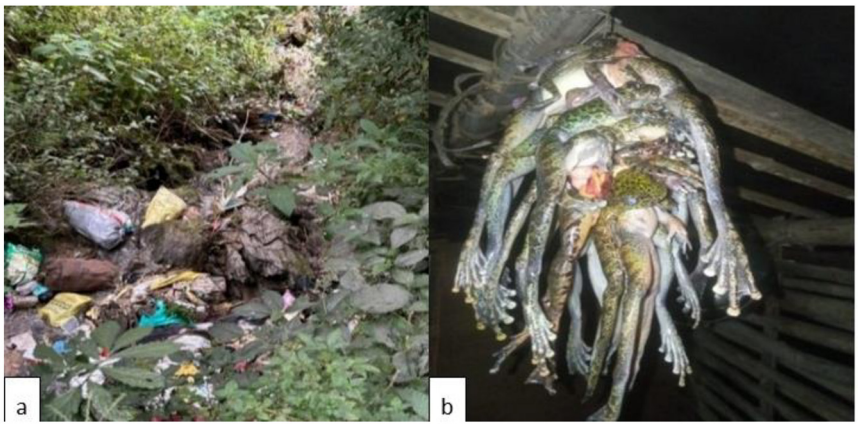



**Fig. 5. (a).** Stream pollution from garbage dumping threatens amphibian microhabitats, **(b).** Hunting of certain amphibian species, such as Amolops sp., by locals is driving their population decline.

certain human-modified habitats can retain significant biodiversity when structural complexity and resource availability are preserved (Nogueira *et al.* 2021). However, the complete absence of *Euphlyctis cyanophlyctis* and *Polypedates maculatus* from the forest underscores the sensitivity of certain taxa to even low-level forest degradation.

An additional driver of population decline in the region is subsistence hunting of certain amphibian species, including *Amolops* spp., *Nanorana* spp., and *Megophrys* spp. (Fig. 5b), particularly for local delicacy and medicinal uses (Shrestha and Gurung, 2019). In contrast, the tea garden enforces restricted worker oversight, reducing poaching. This anthropogenic filter likely contributes to the greater abundance in the tea garden.

The tea garden, therefore, functions as a local refugium that buffers populations of these resilient generalist species against broader regional declines driven by climate change, habitat loss and fragmentation, overexploitation, poaching, and other anthropogenic pressures elsewhere in the Darjeeling-Sikkim Himalaya, as supported by the matrix quality hypothesis enabling higher abundances through preserved structural complexity, hydrological stability, and reduced poaching (Kattel 2022).

Importantly, the study does not suggest that tea gardens are universally superior to natural forests; rather, they complement forest remnants by supporting higher densities of common species while the forest harbors habitat specialists. The landscape-scale partitioning of diversity underscores the importance of maintaining habitat heterogeneity in the Eastern Himalayas.

## CONCLUSION

This study demonstrates that a well-managed anthropogenic ecosystem can support amphibian species richness and abundance comparable to, or slightly greater than, those of a nearby natural montane forest. However, high diversity does not always equate to high habitat quality and may instead lead to 'ecological traps.' The habitat quality governs amphibian persistence. The tea garden's lower pollution load, characterized by minimal pesticide runoff, retained shade trees, and streams largely free of solid waste, creates a more permissive environment than the degraded forest matrix, where garbage dumping, idol immersion, and subsistence hunting directly degrade water quality and suppress populations of species such as *Amolops* spp. and *Nanorana* spp. These conditions align with the matrix quality hypothesis, enabling the tea garden to function as a local refugium that buffers resilient generalists against regional declines driven by climate change, habitat fragmentation, and overexploitation.

Thus, tea gardens in the Darjeeling region should

not be viewed merely as degraded habitats but as valuable complementary landscapes that contribute to regional biodiversity conservation by buffering resilient generalists against broader anthropogenic pressures, while forests harbor habitat specialists.

Nevertheless, the long-term persistence of shared species in the tea garden remains contingent upon sustainable management. Pesticide runoff and seasonal drainage can degrade breeding habitats, potentially reducing amphibian populations in the future. Therefore, integrated conservation must prioritise pollution abatement, sustainable tourism protocols, and anti-poaching enforcement, alongside climate-adaptive strategies to safeguard amphibian diversity across these human-dominated landscapes.

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