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Diversity and distribution analysis of Myxomycetes across Ecological Gradients of Sikkim Himalaya

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ABSTRACT

Myxomycetes are widely distributed across the globe, inhabiting environments ranging from temperate and tropical forests to deserts and high-altitude ecosystems. The present study assessed the diversity and distribution of myxomycetes across tropical, temperate, and sub-alpine terrestrial ecological zones of the Sikkim Himalaya. A year-long field survey recorded 61 specimen (plasmodial and sporocarp) samples representing 6 families, 13 genera, and 21 species. Most collections were obtained post-monsoon season from decaying wood, bark, and leaf litter, highlighting their role as decomposers and indicators of forest health. The Diversity indices like Shannon, Simpson, and Evenness, were highest in the temperate zone, followed by the tropical and sub-alpine zones, indicating altitudinal, substrate availability and microclimatic effects on diversity. Trichiaceae and Stemonitidaceae emerged as dominant families, with *Hemitrichia*, *Arcyria*, and *Stemonitis* as the most frequently encountered genera. This baseline survey data provides new insights into the biogeography of myxomycetes in the Eastern Himalayan region, Sikkim.

Keywords: Myxomycetes, Slime molds, Sikkim Himalaya, Ecological zones, Diversity indices

INTRODUCTION

Myxomycetes, or plasmodial slime molds were initially classified under the phylum Protozoa due to their amoeboid characteristics and phagocytic feeding, they are now placed within the Amoebozoa clade, which includes both slime molds and amoeboid protists (Stephenson, 2011). They thrive on decaying wood, bark, and leaf litter, making them excellent indicators of forest health and ecosystem integrity (Stephenson, Donno, and Schnittler, 2011). They feed on bacteria, yeasts, and other microorganisms and play an important role in balancing soil microbial communities in forest ecosystem (Novozhilov *et al.,* 2018). Scientific interest in Myxomycetes has expanded significantly due to their ecological, biochemical, and biotechnological importance (**Baba, and Sevindik,** 2018; Tran and Adamatzky, 2017; Stoyneva-Gärtner, 2023; Tran *et al.,* 2012). Despite lacking a nervous system, myxomycetes exhibit chemotaxis and problem-solving abilities, which have inspired research in fields such as transport system modeling and urban planning (Bonner, 2010; Tomoru, Hiroki, and Yasuhiko, 2021).

In India, myxomycete research has a long history, beginning with the foundational works of Lister (1924), Lodhi (1934), and Thind (1977), Agnihothrudu (1961), followed by monographic treatments such as that of Lakhanpal and Mukerji (1981) and comprehensive checklists by Ranade *et al.,* (2012), which together document more than 450 species. Sikkim, a recognized biodiversity hotspot in the Eastern Himalayas, offers ideal conditions for myxomycete growth due to its high rainfall, diverse forest types, and abundant decaying organic matter. Considering the ecological significance and bioactive potential of myxomycetes, and the scarcity of regional data from the Sikkim Himalaya, the present study aims to investigate their diversity and distribution across major ecological zones. The study will provide essential baseline information to enhance regional understanding of myxomycetes diversity in Sikkim.

MATERIALS AND METHODS

Study Area and Sampling

The study was conducted across three major ecological zones of Sikkim: tropical-subtropical (300–1200 m), temperate (1200–3000 m), and sub alpine to alpine (above 3000 m) **(Figure 1).**

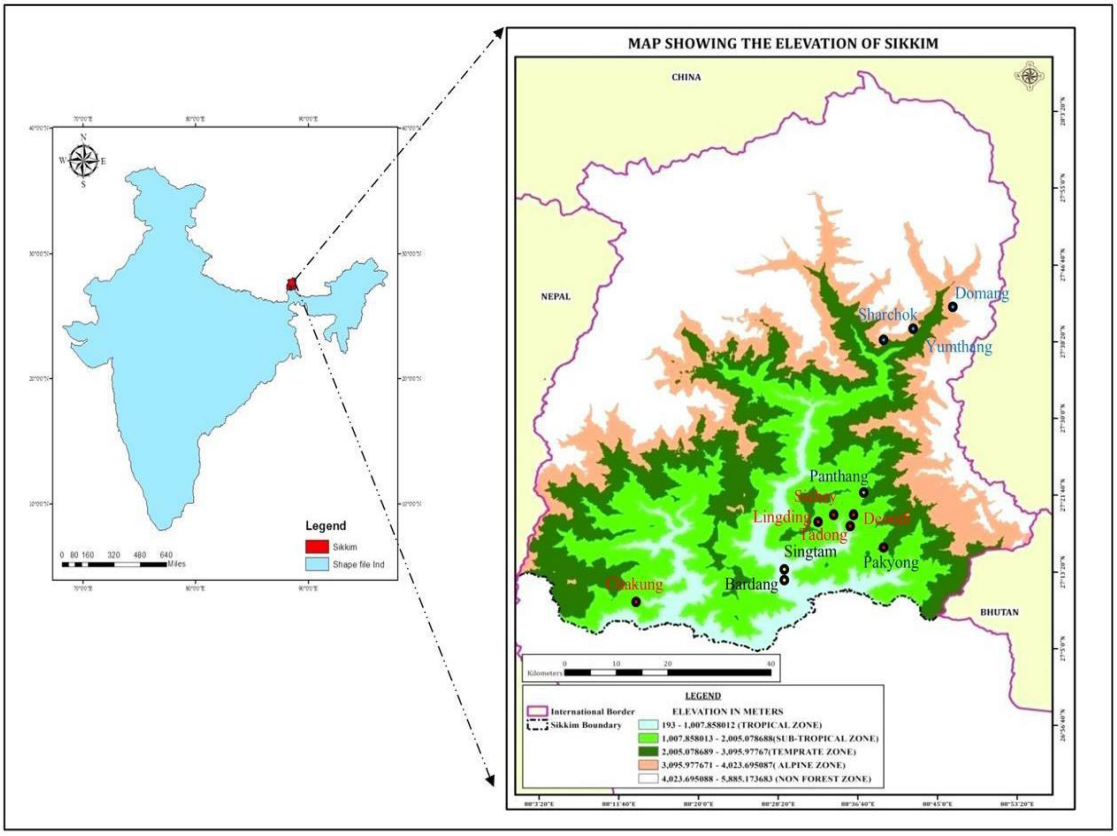


Figure 1. Map of study site Sikkim, highlighting the three ecological zones.

These zones were selected based on climatic distinctiveness, vegetation composition, and accessibility. A 10× hand lens aided in spotting small specimens, and GPS-enabled mobile devices recorded coordinates and elevation. Macro- and microhabitat photographs were taken using a Nikon digital camera and Samsung S23. Field sampling, conducted from August 2024 to July 2025. Habitat and substrate characteristics (e.g., wood, bark, leaf litter) were also recorded to aid species identification. The morphological characteristic were recorded in their natural habitat. Collected specimens were placed in labelled Petri dishes or containers and brought to the lab for morphological and culture analysis (Basanta and Torres 2017).

Morphological Characterization and Identification

Microscopic and macroscopic examinations of the specimens were carried out using a stereo zoom microscope. Identification was carried out using a combination of macroscopic and microscopic diagnostic characters, including fruiting body type (sporangia, aethalia, plasmodiocarp), size, colour, stalk characteristics, and capillitium structure. Substrate type and habitat conditions were also considered as supporting criteria. In cases where only the plasmodial stage was observed and fruiting bodies were absent; identification was restricted to the genus level to ensure taxonomic accuracy.

Key morphological characteristics were documented, including: **(i)** *Fruiting body type*—such as sporangia (stalked or sessile), aethalia, pseudoaethalia, and plasmodiocarps; **(ii)** *Peridium and stalk*—texture, coloration, and ornamentation of the outer wall and supporting stalks; **(iii)** *Capillitium*—structure, branching pattern, pigmentation, and the presence of lime deposits, particularly important for members of Physarales; and **(iv)** *Plasmodium*—when present, its color and movement.

Taxonomic identification of the collected myxomycete specimens was conducted using standard dichotomous keys and species descriptions provided in *The Myxomycetes* by Martin and Alexopoulos (1969), Thind (1977), Lakhanpal and Mukherji (1981), Stephenson and Stempen (1994), Ing and Stephenson (2022), Lado, (2021), Li *et al.,* (2008). Additional online databases, including Nomen.eumycetozoa.com, Index Fungorum, and the Indian Myxomycetes Database were also referred to validate the identification.

Data Analysis

Quantitative diversity indices were computed to assess species richness and community composition across zones. The following indices were used: **Shannon Diversity Index (H′)** to evaluate richness and evenness, **Simpson's Diversity Index (1 − D)** to assess species dominance, **Evenness Index (E_h)** to understand community balance. Statistical analysis was performed using Microsoft Excel.

RESULTS

Distribution of myxomycetes Family and Genera

A total of 61 myxomycete specimens were recorded across the three ecological zones of Sikkim—Tropical- subtropical (18), Temperate (36), and Sub-alpine/Alpine (7). The most dominant family was Trichiaceae (17 occurrences), followed by Stemonitidaceae (15), Arcyriaceae (10), Physaraceae (8), Cribrariaceae and Tubiferaceae (3 each) and Ceratiomyxaceae (3). Didymiaceae and Dictydiaceae with one record each. The genus *Hemitrichia* was the most frequently observed with 13 occurrences, followed by *Arcyria* (10), *Stemonitis* (9), and *Lycogala (*3). *Physarum(*3)., *Ceratiomyxa*, *(*3), *Cribraria(*3), *Trichia* and *Dictydium* (2), while *Metatrichia, Fuligo, Comatricha,* and *Didymium* with single record. These genera were distributed across ecological zones, reflecting the adaptability of Myxomycetes to diverse environmental conditions and highlighting the region's rich yet underexplored myxomycete diversity. A total of 21 species were recorded in the present study as summarized in **Table 1, 2, 3**. This diversity underscores the ecological complexity and favourable microhabitats present across the altitudinal gradient of Sikkim Himalayas.

The tropical region of Sikkim has a warm climate with moderate to heavy monsoon rainfall (June–September) and temperatures from 13–30°C. It features lush, dense forests with tropical hardwoods, ferns, and shrubs. The humid conditions and abundant organic matter on the forest floor provide ideal habitats for myxomycetes with *Hemitrichia*, *Arcyria* and *Physarum* frequently sighted genera **(Table 1, Figure 2**).

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Table 1. Morphological characterization of myxomycetes of the tropical & subtropical zone of Sikkim Himalaya.

S.No.	Species Name	Family	Order	Key diagnostic characters	Size	Substrate	Season
1	*Hemitrichia serpula* (Scop.) Rostaf. ex Lister, 1894	Trichiaceae	Trichiales	Serpentine, coiled plasmodiocarp,orange to yellow;well-developed capillitum	1.4 cm	Dead wood	Autumn, Summer
2	*Metatrichia vesparium* (Batsch) Nann.-Bremek. ex G.W. Martin &Alexop.	Trichiaceae	Trichiales	Short-stalked to sessile reddish-brown sporangia, dense capillitium	0.5 cm	Dead wood	Autumn
3	*Arcyria denudata* (L.) Wettst., 1886	Arcyriaceae	Trichiales	Tall, slender stalks with cream globose sporangium; exposed capillitum	0.4 cm	Dead wood	Autumn
4	*Arcyria cinerea* (Bull.) Pers., 1801	Arcyriaceae	Trichiales	Pale gray cylindrical sporangia, exposed capillitium	—	Decaying wood	Autumn, Summer
5	*Cribrariacancellata* (Batsch) Nann.-Bremek., 1975	Cribrariaceae	Cribrariales	Netted sporangia, lattice-like structure on stalks	—	Decaying wood, bark	Autumn, Summer
6	*Physarum polycephalum* Schwein., 1822	Physaraceae	Physarales	Bright yellow veiny plasmodium, clustered sporangia	—	Decaying wood	Autumn
7	*Stemonitis splendens*Rostaf., 1874	Stemonitidaceae	Stemonitales	Tall dark cylindrical sporangia on slender stalks, fibrous capillitium	1.0 cm	Decaying wood	Autumn, Summer

8	*Stemonitisfusca* Roth, 1787	Stemonitidaceae	Stemonitales	Tall, dark cylindrical sporangia, net-like capillitium	—	Rotting wood	Autumn, Summer
9	*Lycogala epidendrum* (L.) Fr., 1829	Tubiferaceae	Liceales	Pink spherical fruiting bodies, soft texture, brown at maturity	0.3 cm	Decaying wood	Autumn
10	*Physarum pezizoideum* (Jungh.) Pavill. & Lagarde, 1903	Physaraceae	Physarales	Cup-shaped sporangia, thin peridium, pale bluish to whitish	0.3 cm	Decaying wood	Autumn
11	*Ceratiomyxa fruticulosa* (O.F. Müll.) T. Macbr., 1899	Ceratiomyxaceae	Ceratiomyxales	White;coral like branching fruiting body with cup-shaped sporangia on tips	1.3 cm	Decaying wood	Summer
12	*Hemitrichia calyculata* (Speg.) M.L. Farr, 1974	Trichiaceae	Trichiales	Small stalked bodies with cup-like apex, orange sporangia	—	Decaying bark	Summer

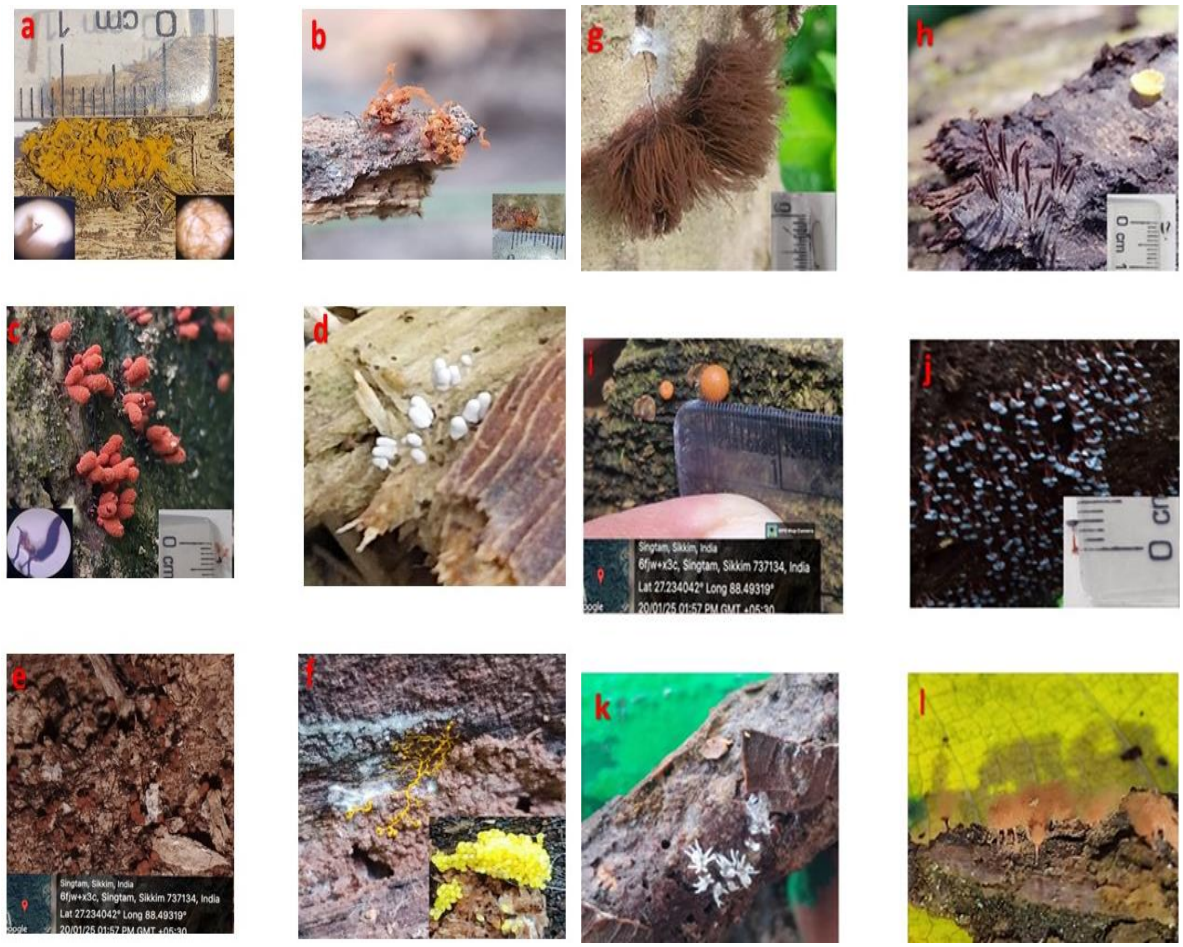


Figure 2 (a-l). Myxomycete species recorded from the Tropical zone: (a)Plasmodiocarp of *Hemitrichia serpula* (b) Sporocarp of *Metatrichia vesparium* (c) Sporocarp of *Arcyria denudata* (d) Sporocarp of *Arcyria cinerea* (e) Sporocarp of *Cribraria cancellatum* (f) Planeroplasmodium and immature fruiting body of *Physarum polycephalum* (g) Sporocarp of *Stemonitis splendens* (h) *Sporocarp* of *Stemonitis fusca* (i) Immature aethelium of *Lycogala epidendrum* (j) Sporocarp of *Physarum pezizoideum* (k) Sporocarp of *Ceratiomyxa fruticulosa* (l) Sporocarp of *Hemitrichia calyculata.*

The temperate zone of Sikkim has cooler temperatures (10–20°C), distinct seasons, and moderate year-round rainfall. Its forests include broadleaf and coniferous trees, along with abundant mosses and lichens. The region supports rich biodiversity and shows greater seasonal variability than the tropical zone. These conditions favour myxomycetes development, especially during the monsoon and post-monsoon periods, with genera such as *Hemitrichia*, *Arcyria*, and *Stemonitis* were frequently recorded **(Table 2, Figure 3).**

Table 2. Morphological characterization of myxomycetes of the temperate zone of Sikkim Himalaya.

S.No	Species Name	Family	Order	Key diagnostic characters	Substrate	Season(s)
1	*Arcyria denudata* (L.) Wettst.	Arcyriaceae	Trichiales	Cream to pale brown stalked sporangia;globoseheads;exposed capillitium;texturesoft,cottony	Dead wood	Winter, Spring, Summer, Autumn
2	*Stemonitis splendens*Rostaf., 1874	Stemonitidaceae	Stemonitales	Tall dark brown/black stalks; cylindrical sporangia; dense fibrous capillitium	Decaying wood	Winter, Spring, Summer
3	*Hemitrichiaserpula* (Scop.) Rostaf. ex Lister	Trichiaceae	Trichiales	Serpentine, coiled fruiting bodies; orange to yellow-orange	Dead wood	Winter, Spring, Summer
4	*Hemitrichiacalyculata* (Speg.) M.L.Farr	Trichiaceae	Trichiales	Stalked, cup-like apex; orange to yellow-orange sporangium	Decaying bark	Winter, Spring, Summer
5	*Dictydium cancellatum* (Batsch) T. Macbr.	Reticulariaceae	Liceales	Stalked fruiting bodies with net-like (reticulate)sporangium	Decaying wood	Spring
6	*Trichia decipiens* (Pers.) T. Macbr.	Trichiaceae	Trichiales	White plasmodium turns pink/red; olive/yellow-brown conical sporangia at maturity	Decaying wood, bark	Spring, Summer
7	*Fuligoseptica* (L.) F.H. Wigg., 1780	Physaraceae	Physarales	"Dog vomit" mold; yellow/orange plasmodium; sponge-like aethalium	Decaying wood	Spring
8	*Physarum viride* (Bull.) Pers., 1795	Physaraceae	Physarales	Greenish/olive sporangia, mostly sessile	Decaying wood	Spring
9	*Lycogala epidendrum* (L.) Fr., 1829	Tubiferaceae	Liceales	"Wolf's milk"; pink to brown ovoid fruiting bodies in clusters	Decaying wood	Winter, Summer
10	*Ceratiomyxafruticulosa* (O.F. Müll.) T. Macbr.	Ceratiomyxidae	Protosteliales	Tree-like fruiting bodies with cup-like sporangia on branch tips	Decaying wood	Winter, Summer
11	*Arcyria cinerea* (Bull.) Pers., 1801	Arcyriaceae	Trichiales	Pale gray, cylindrical clustered sporangia; exposed capillitial threads; texture powdery	Decaying wood	Summer
12	*Stemonitisfusca* Roth, 1787	Stemonitidaceae	Stemonitales	Tall, blackish cylindrical sporangia; net-like capillitium	Rotting wood	Summer
13	*Arcyriaobvelata*(Oeder) Onsberg, 1979	Arcyriaceae	Trichiales	Pink/red cylindrical sporangia with flaking peridium; thread-like spiralled capillitium; texture soft, fluffy	Decaying wood, bark	Summer
14	*Stemonitisaxifera* (Bull.) T. Macbr., 1889	Stemonitidaceae	Stemonitales	Slender brown cylindrical sporangia in clusters; net-like capillitium	Rotting wood	Summer
15	*Cribraria violacea* Rex, 1893	Cribrariaceae	Cribrariales	Violet, netted sporangia; reduced peridium with dictydine granules; delicate stalk	Decaying wood	Summer

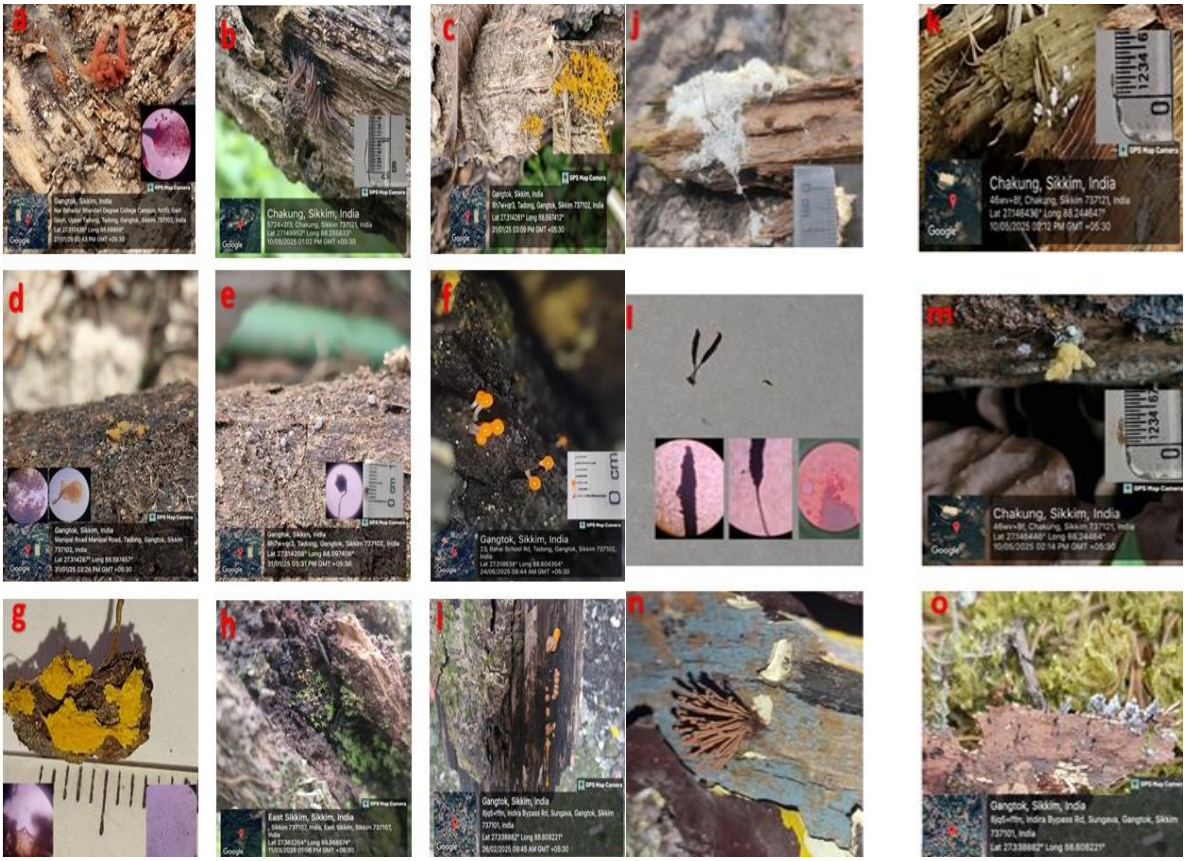


Figure 3 (a-o). Myxomycete species recorded from the Temperate zone: (a) Sporocarp of *Arcyria denudata* (b) Sporocarp of *Stemonitis splendens* (c)Plasmodiocarp of *Hemitrichia serpula* (d)Sporocarp of *Hemitrichia calyculata* (e)Sporocarp of *Dictydium cancellatum* (f) Sporocarp of *Trichia decipiens* (g) Aethalium of *Fuligo septa* (h)Sporocarp of *Physarum viride* (i) Aethelium of *Lycogala epidendrum* (j) Sporocarp of *Ceratiomyxa fruticulosa* (k) Sporocarp of *Arcyria cinerea* (l) Sporocarp of *Stemonitis fusca* (m) *Sporocarp of Arcyria oblevata* (n) Sporangium of *Stemonitis axifera* (o) Sporocarp of *Cribraria violacea.*

The alpine and sub alpine zone of Sikkim experiences cold conditions, with temperatures from –5°C to 10°C and heavy winter snowfall. Summers are short and cool, with a dry period during the colder months. Vegetation is sparse, dominated by alpine meadows, hardy grasses, and small high-altitude plants. Despite the harsh environment, some myxomycetes recorded, especially in sites with accumulated organic matter. Their presence at these elevations reflects their ability to adapt to diverse ecological niches across Sikkim **(Table 3, Figure 4).**

Table 3. Morphological characterization of myxomycetes of the Alpine zone of Sikkim Himalaya.

S.No.	Species Name	Order	Family	Key diagnostic characters	Substrate	Season
1	*Lycogala epidendrum* (J.C. Buxb. ex L.) Fr. 1829	Liceales	Tubiferaceae	Small, spherical to ovoid fruiting bodies; pink when immature, turning brown at maturity; often occurring in clusters;soft texture	Decaying wood	Autumn, Spring
2	*Physarum viride* (Bull.) Pers., 1795	Physarales	Physaraceae	Greenish to olive-coloredsporangia;typically sessile or with short stalks	Decaying wood	Autumn
3	*Comatricha tenerrima* (M.A.Curtis) G.Lister, 1919	Stemonitales	Stemonitidaceae	Very small, black, long-stalked sporangia; delicate and slender	Decaying wood	Autumn
4	*Hemitrichiacalyculata* (Speg.) M.L.Farr, 1974	Trichiales	Trichiaceae	Stalked sporangia with distinct cup shaped(calyculate) apex; orange to yellow-orange; well-developed capillitium	Decaying bark of tree	Winter,Spring
5	*Didymium nigripes* (Link) Fr., 1829	Physarales	Didymiaceae	Shiny, dark sporangia with a black stalk	Decaying wood	Spring



Figure 4 (a-e). Myxomycetes species recorded from the Alpine and subalpine Zone: (a) Aethalium- light pink immature and grey mature of *Lycogala epidendrum* (b) Sporocarp of *Physarum viride* (c) Sporocarp of *Hemitrichia calyculata* (d) Sporocarp *of Comatricha teledrama* (e) Sporocarp of *Didymium nigripes.*

Diversity Indices

Quantitative analysis using diversity indices further revealed significant differences among the three zones. These metrics suggest that the temperate zone harbors a more stable and ecologically diverse myxomycete community, possibly due to favorable microclimatic conditions and greater habitat heterogeneity. The key values of each index are summarized in **Table 4.**

Table 4. Diversity Indices of Myxomycetes across ecological Zones in the Sikkim Himalaya.

Parameter	**Tropical – subtropical Zone**	**Temperate Zone**	**Alpine – subalpine Zone**
Species Richness (S)	12	15	5
Shannon Diversity Index (H′)	2.399204	2.529761	1.549826
Simpson's Diversity Index (1 – D)	0.9542	0.9365	0.9048
Species Evenness (E_h)	0.979	0.986	0.963

Seasonal Fruiting Patterns

A clear pattern of seasonal fruiting was observed, post-monsoon months (September–December) yielding the highest number of fruiting bodies. This corresponds with increased moisture availability and favorable microclimatic conditions. In contrast, pre-monsoon surveys recorded fewer specimens and limited sporulation.

DISCUSSION

The present study attempts to provide a detailed distribution and ecological analysis of myxomycete across three ecological zones; tropical, temperate, and alpine in the Sikkim Himalaya. A total of 61 specimens (plasmodial and sporocraphs) representing 21 species were recorded, with the temperate zone with highest species richness of 15, followed by the tropical 12 and alpine 5 zones. This pattern is consistent with previous studies from Northeast India, where mid-altitudinal regions are recognized as favorable habitats for myxomycetes due to optimal environmental conditions (Sharma and Lakhanpal, 2023).

The distribution data obtained within a short sampling period and limited surveys especially in alpine and tropical regions reinforce the Eastern Himalayas emerging status as a hotspot of slime mold diversity. The Shannon Diversity Index (H′) revealed that the temperate zone had the highest species diversity (H′ = 2.529761), followed by the tropical-subtropical zone (H′ = 2.399204) and alpine-subalpine zone (H′ = 1.549826). These values suggest that the temperate region supports a more diverse and balanced myxomycete community, while the tropical zone, although species-rich, shows slightly lower diversity. The Simpson's Diversity Index (1 − D) followed a similar trend, with values of 0.9365 for temperate, 0.9542 for tropical, and 0.9048 for alpine zones, indicating low dominance by any single species across all zones and overall ecological stability. Species evenness (E_h) was highest in the temperate zone (0.986), followed by the tropical-subtropical (0.979) and alpine-subalpine zones (0.963), indicating relatively uniform distribution of individuals across species in these regions. The high evenness in all three zones reflects a balanced species structure with minimal dominance and suggests stable ecological niches (Ing and Stephenson, 2022). The observed altitudinal variation in species distribution can be attributed to differences in microclimatic factors such as temperature, humidity, and substrate availability. Substrate preference was evident, with most species collected from dead-decaying wood, tree bark, and rotting wood - substrates known to provide high organic content and moisture retention crucial for slime mold development. This variation in richness across altitudes highlights the role of climatic and ecological heterogeneity in shaping their diversity.

The higher diversity in the temperate zone aligns with findings that moderate climates offer optimal temperature and moisture for microbial proliferation. Conversely, the relatively lower richness and diversity in the alpine zone likely reflect the environmental constraints imposed by cold temperatures, limited substrate availability, and shorter growing seasons (Rojas and Stephenson, 2008). Seasonal patterns were evident, with post-monsoon conditions enhancing fruit body development, aligning with findings of Chen *et al.,* (2024). Compared to previous myxomycete studies from Northeast India, which has largely focused on taxonomic inventories (Sharma and Lakhanpal, 2023), the present work integrates ecological parameters, altitudinal gradients, and seasonal influences with quantitative diversity indices, providing a more holistic view of myxomycete distribution. The detection of rarely reported species such as *Physarum polycephalum* and high-altitude taxa such as *Comatricha tenerrima,* and *Didymium nigripes* adds important biogeographical insights to the limited myxomycete data available from the Indian Himalayas.

Despite the limitations such as restricted sampling periods and access to some microhabitats, the present study demonstrates that altitudinal gradient, substrate availability, and seasonal moisture are major determinants of Myxomycete community structure in Sikkim Himalaya. The temperate zone acts as a biodiversity hotspot, while tropical and alpine zones harbor distinct assemblages adapted to their respective environmental conditions. Integrating ecological, climatic, and microhabitat variables is essential for advancing our understanding of myxomycete ecology in Himalayan ecosystems. The detection of rarely reported taxa and the observation of plasmodial stages in certain species highlight both the diversity

and the limitations of morphology-based identification, thereby supporting the need for incorporating molecular approaches in future studies.

CONCLUSION

The present study documents the diversity of myxomycetes across three ecological zones of Sikkim Himalaya, and the temperate zone emerged as the most species-rich, likely due to its favorable microclimatic conditions and substrate availability. Peak fruiting during the post-monsoon season underscores the importance of moisture availability, while the consistent occurrence of species on decaying wood substrate highlights their role as key decomposers within forest ecosystems. Quantitative diversity indices indicate a well-structured and balanced myxomycete community. The detection of rare high-altitude taxa further reflects the ecological resilience of certain species and emphasizes the need for continued surveys in underexplored zones. While this study provides an essential baseline for understanding myxomycete distribution and also highlights the integration of ecological, climatic, and microhabitat variables is essential for developing a holistic understanding of myxomycete distribution patterns in the Sikkim Himalayan region. It also highlights the need for further investigations incorporating molecular tools and expanded sampling strategies to better understand species diversity and ecological dynamics.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest regarding the publication of this work.

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