

DAVIS EXPEDITION FUND

REPORT ON EXPEDITION / PROJECT

Expedition/Project Title:	Lichen Diversity in the Himalayan Forest Ecosystems: Understanding Their Applicability for Nitrogen Pollution Bioindication
Travel Dates:	31 August – 14 December 2025
Location:	Lalitpur, Makwanpur, Dhading, Ramechhap, Rasuwa, Sindhupalchok and Solukhumbu Districts of Nepal
Group Members:	Suman Prakash Pradhan, Sujan Prakash Pradhan, Asmita Shrestha and others (see details below)
Aims:	Sampling of lichen community structure, collection of lichen specimens for taxonomic/molecular identification, collection of lichen samples for chemical analyses, and deploying the ALPHA passive sampler for monitoring atmospheric ammonia
Photography consent form attached: (please refer to your award letter)	<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No

Summary

15-week fieldwork in nine different sites of Central Nepal for the sampling of lichen community structures, collection of lichen specimens for taxonomic/molecular identification, collection of lichen samples for chemical analyses, and deploying ALPHA passive samplers for atmospheric ammonia monitoring was accomplished from 31st August to 14th December 2025.

A total of 3238 specimens of lichens, along with 81 selective samples for chemical analyses, were collected and brought to the Royal Botanic Garden Edinburgh (RBGE). An additional 3238 duplicate reference specimens were deposited at the National Herbarium and Plant Laboratories (KATH) of the Ministry of Forests and Environment, Government of Nepal. Moreover, the triplicates of ALPHA passive samplers in three different elevations of a site were deployed in all nine sites to monitor average atmospheric ammonia concentrations over two-month periods for a total of 12 months. The field measurement data for atmospheric ammonia concentration will be utilised to ground-truth modelled data and also to correlate them with lichen responses to levels of nitrogen pollution. Further, during the same fieldwork, a biosocial survey on perceptions of indigenous people and local communities on nitrogen air pollution and lichen bioindicators was conducted with 20 respondents in each site.

The collaboration with the Department of Plant Resources (DPR), Government of Nepal and the Kathmandu University, was established for continuation of this work and future research.

Currently, all specimens are being identified morphologically at RBGE, and the chemical analyses for tissue carbon, nitrogen and isotopic signature are being carried out at the University of Edinburgh. The laboratory analyses for the ALPHA samplers are being carried out at the UK Centre for Ecology & Hydrology, Edinburgh.

Detailed Report

1. Background

Lichens are non-vascular cryptogams and obligatory ecological symbioses between a heterotrophic mycobiont (fungi), and an autotrophic photobiont (green algae and/or cyanobacteria). Furthermore, bacteria, viruses, endophytic and lichenicolous fungi represent additional components of the lichen microbiome. Lichens are widely distributed from the arctic to the tropics, and from the plains to the highest mountains, and live in habitats from rivers to deserts (Jha et al., 2017). Lichens are commonly known as "*Jhyau*" in Nepali, and their distribution covers the Tarai lowlands to the Himalayan highlands. Wallich's collection, which took place between 1820 and 1826, was the first to generate knowledge of lichens in Nepal (Thapa and Rajbhandary, 2012). Subsequently, additional publications were focused on the taxonomic examination of specific genera, or the lichen flora found in either a specific region or the whole country. Even today, the lichens found in the lowland Tarai and Siwalik highlands of Nepal are not well documented, and those from the western region of Nepal are essentially unknown (Bhujju et al., 2007). To date, a total of 873 species of lichen-forming fungi have been identified, divided into 185 genera (Baniya and Bhatta, 2021), with many more still to be explored and identified.

Lichens, which rely almost entirely on wet and dry atmospheric deposition for their nutritional needs, are sensitive to environmental change, particularly air pollution (Munzi et al., 2019). Despite often having a wider distribution than vascular plants and other cryptogams, such as bryophytes, lichen species are among the most vulnerable organisms to alterations in the physical environment (Saipunkaew et al., 2007). Lichens can absorb nitrogen and other elements into their thalli; the abundances of which, can reflect the concentration of those pollutants in the surrounding environment. The concentration of atmospheric ammonia and nitrogen deposition above which there are negative impacts on the physiology of lichens are defined as an ammonia 'critical level' and nitrogen 'critical load'. Those threshold concentrations – if they can be identified for the Himalayan forest ecosystems – will be a first step towards understanding the risk of nitrogen pollution in the Himalayas. A recent desk-based meta-analysis warned that ca. 90-95% Himalayan forest ecosystems currently exceeded the ammonia critical level and nitrogen critical load, while cautioning that there remained an urgency for more field-based research to clarify real-world impacts (Delves et al., 2023; Ellis et al., 2022).

In recent years, research on lichen epiphytes as biological indicators has started to gain some attention in Nepal (Pradhan et al., 2026; Pradhan et al., 2025). Nonetheless, the application of lichens in nitrogen pollution biomonitoring needs additional resourcing. A change in community structure due to differential sensitivity/tolerance of lichens with contrasting functional traits is one of the potential primary indications of nitrogen stress (Johansson et al., 2012). In general, the disappearance of sensitive lichens from a particular habitat is an outcome of increased nitrogen concentration in the atmosphere. Thus, the characterisation of community structure, abundance and diversity of Nepalese lichens, constituting different functional groups, and assigning these into bioindicator categories such as oligotrophic, nitrophytic and mesotrophic, is proposed to enhance monitoring and management of nitrogen pollution in the Himalayas.

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This study aimed to monitor nitrogen air pollution and systematically sample lichens from deciduous forests of the temperate climatic zone of central Nepal – sites positioned along nitrogen gradients, with the following specific objectives.

- i) To systematically sample lichens from deciduous forest sites in the temperate climatic zone of central Nepal.
- ii) To analyse the diversity and functional traits of lichens, and compare these to the level of atmospheric ammonia concentration.
- iii) To demonstrate the source of nitrogen in the different sites, with lichen epiphytes as indicators.
- iv) To advance taxonomic works on lichen epiphytes from the Nepal Himalayas.
- v) To conduct a biosocial survey to document perceptions of indigenous people and local communities on nitrogen pollution and lichen bioindicators.

The 15-week-long field work has now been completed with the collection of lichen specimens and samples, deploying ALPHA passive samplers and conducting biosocial surveys from 31st August to 14th December 2025.

2. Methods

2.1. Sites

The field work was carried out in nine deciduous forest sites in the temperate climatic zone with an elevation range of 2000-3000 metres above sea level (m a.s.l.), with these being grouped into three zones corresponding to atmospheric ammonia concentration classes of less than/equal to $2.5 \mu\text{g}/\text{m}^3$, between 2.5 and $5 \mu\text{g}/\text{m}^3$ and above $5 \mu\text{g}/\text{m}^3$. These ammonia concentration classes were acquired from the EMEP model outputs with 2010 emission inventory and 2018 meteorology (Ellis et al., 2022). A total of nine sites were selected for lichen collection, three in each of the three pollution classes (Table 1 & Figures 1, 2, 3). These sites were selected based on the distribution of deciduous forests (which standardises for the broad forest type) and within the temperate climatic zone (which standardises for the broad climate) to minimise confounding effects of vegetation type and climate.

2.2. Sample Collection

Nine trees (three in each of three sampling lines) in a site were selected based on the predetermined criteria such as tree species, DBH, height, and crown cover. The collection of lichen specimens was carried out from all four aspects of a tree by placing a ladder quadrat of 50 cm * 10 cm, divided into five equal sub-quadrates of 10 cm * 10 cm. The lichen species in each sub-quadrant were collected and safely stored in paper envelopes, and high-quality photographs were taken. For the chemical analyses, a total of 81 bulk samples (approximately 10 g) of *Hypotrachyna* spp., *Ramalina* spp., *Usnea* spp., *Parmotrema* spp., *Hypogymnia* spp., *Lobaria* spp., and *Sulcaria* spp. were collected from the same tree. All samples were air-dried, and export permits from the DPR were taken before transporting them to RBGE.

2.3. Atmospheric Ammonia Monitoring

A total of 27 stations for atmospheric ammonia monitoring by UKCEH's Adapted Low-cost Passive

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High Absorption (ALPHA) passive samplers were established. A site consists of three stations with triplicate sets of ALPHAs. Bimonthly monitoring will be carried out until December 2026.

Table 1: Summary of field sites.

Ammonia Concentration Class	Site	Place	Coordinates	Elevation (m a.s.l.)	Vegetation
≤2.5 µg/m ³	S1	Golzong, Rasuwa	28°09'29.10"N 85°17'35.17"E	2456- 2817	Rhododendron, Quercus, Alnus, Pinus
	S2	Phaplu, Solukhumbu	27°32'37.04"N 86°34'01.47"E	2626- 2975	Rhododendron, Quercus, Alnus, Pinus
	S3	Likhupike, Solukhumbu	27°32'53.59"N 86°24'37.59"E	2795- 2964	Rhododendron, Quercus
>2.5 ≤5 µg/m ³	S4	Mude, Sindhupalchok	27°41'07.19"N 85°55'39.03"E	2381- 2611	Rhododendron, Quercus, Alnus, Pinus
	S5	Singatee, Ramechhap	27°32'58.62"N 86°16'26.41"E	2684- 2848	Rhododendron, Quercus
	S6	Pattale, Solukhumbu	27°23'17.78"N 86°32'56.22"E	2709- 2959	Rhododendron, Quercus, Pinus
>5 µg/m ³	S7	Gotikhel, Lalitpur	27°28'51.34"N 85°22'55.21"E	2033- 2143	Rhododendron, Quercus, Pinus
	S8	Tistung-Palung, Makwanpur	27°37'59.07"N 85°01'50.72"E	2310- 2470	Rhododendron, Quercus, Alnus, Pinus
	S9	Toplang, Dhading	27°40'48.31"N 85°10'47.79"E	2196- 2357	Rhododendron, Quercus, Pinus

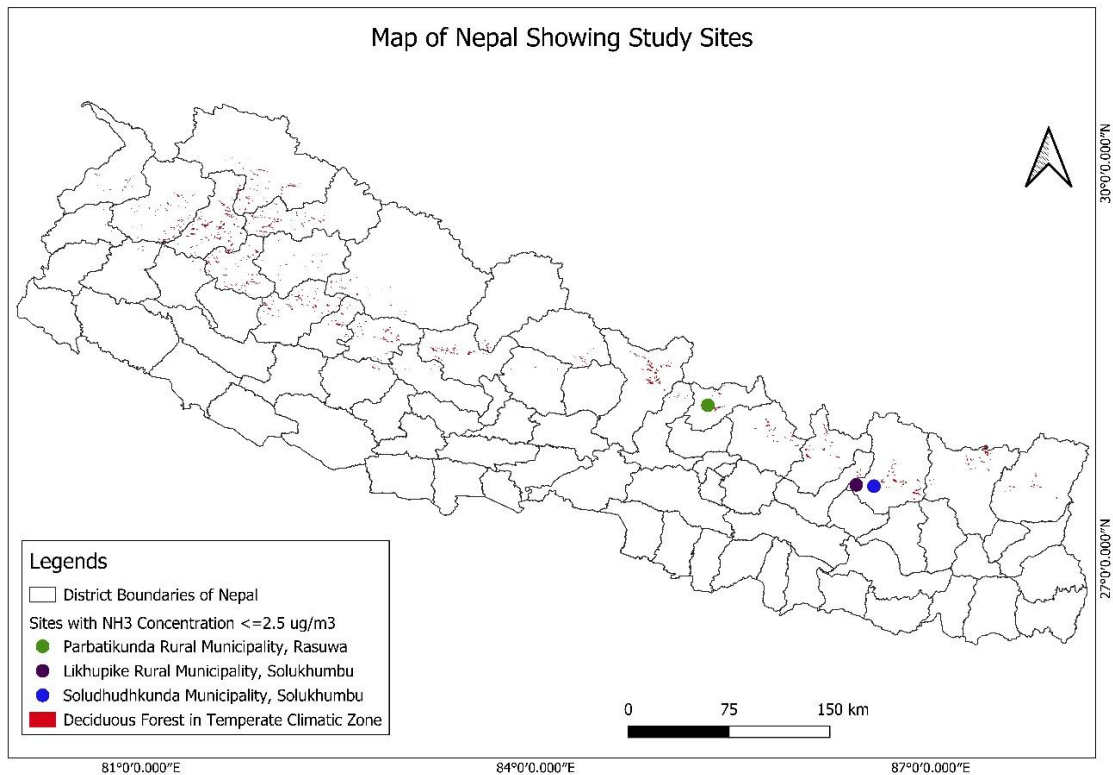


Figure 1: Sites with ammonia concentration below 2.5 µg/m³.

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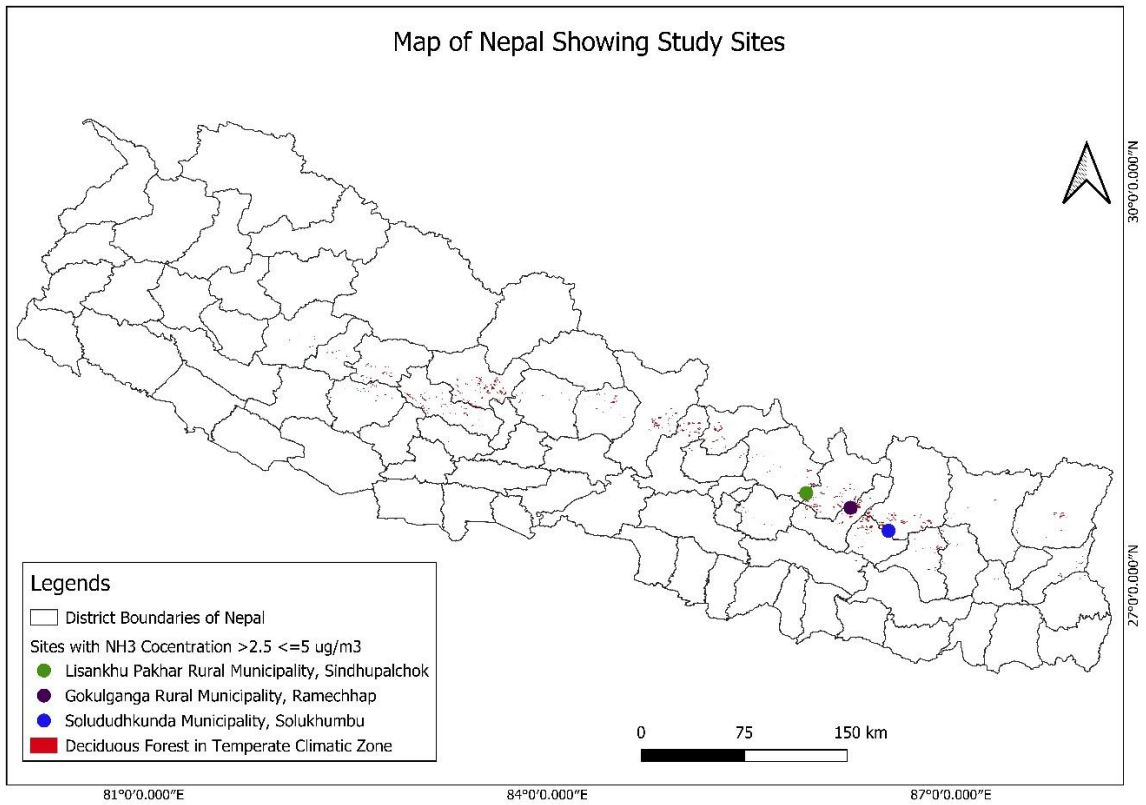


Figure 2: Sites with ammonia concentration above 2.5 and below 5 µg/m³.

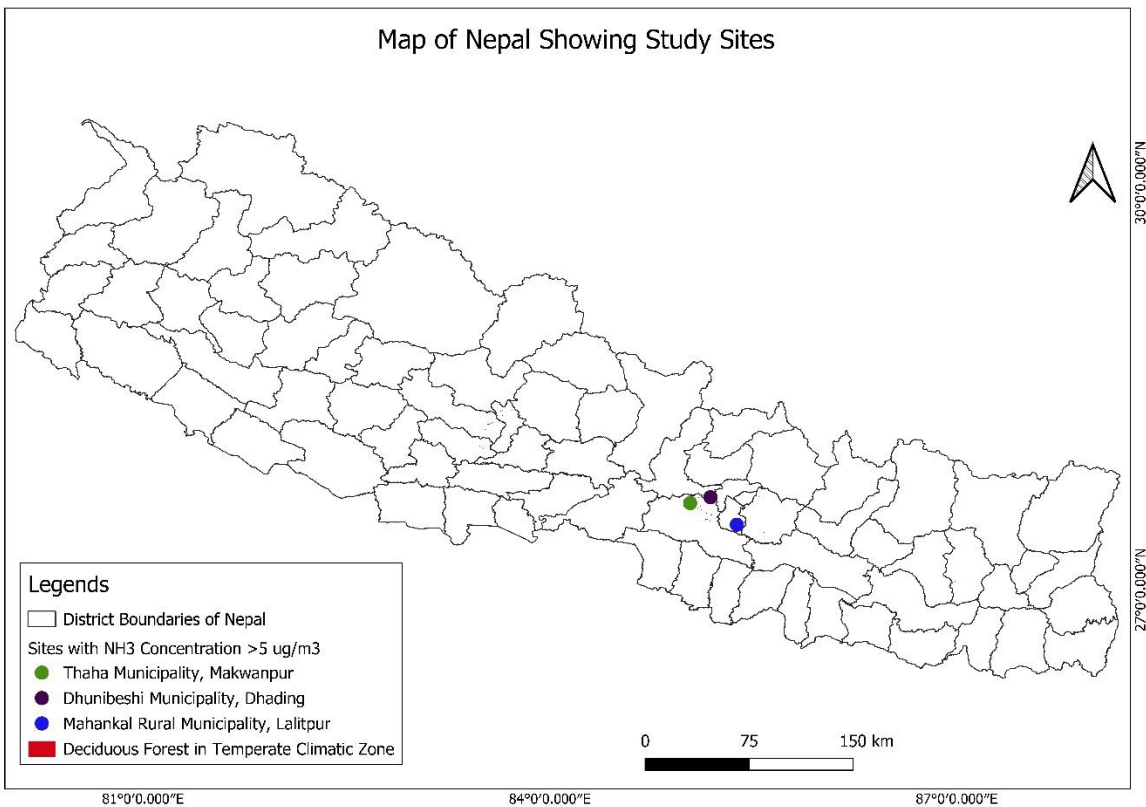


Figure 3: Sites with ammonia concentration above 5 µg/m³.

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3. Accomplished and ongoing works

A total of 3238 lichen specimens (183 from S1, 227 from S2, 241 from S3, 348 from S4, 431 from S5, 382 from S6, 539 from S7, 462 from S8, 425 from S8) were collected and brought to RBGE for taxonomic/molecular identification. The specimens are now being processed to be deposited in the herbarium. The same number of duplicate specimens is deposited at the National Herbarium and Plant Laboratories of Nepal. Additional collection for chemical analyses (81 total samples from nine sites) has been completed, and ALPHA samplers have been deployed for atmospheric ammonia monitoring every two months. The taxonomic identification of lichen specimens is being carried out at RBGE, and chemical analyses for nitrogen, carbon and isotopic signature are being carried out at the University of Edinburgh. The analyses for atmospheric ammonia in ALPHA samplers are being carried out at the UK Centre for Ecology & Hydrology. The details of work and their status are presented in Table 2.

Table 2: Details of accomplished and ongoing works

S.N.	Activities	Objective(s)	Expected outputs	Status	Remarks
1	Collection of lichen specimens	Taxonomic identification and trait analyses	The community structure and different functional traits reflect the stress of nitrogen pollution in lichen bioindicators of the Himalayan forest ecosystems	Collection completed and analyses ongoing	RBGE and Natural History Museum, London
2	Collection of selective lichen samples	Thallus nitrogen, carbon and nitrogen isotopic signature analyses	This reflects the magnitude and sources of nitrogen pollution in the Himalayan forest habitats and will serve as a reference for the constituents of carbon and nitrogen in natural ecosystems	Collection completed and analyses ongoing	University of Edinburgh
3	Collection of tree bark	pH analysis	Suitability of phorophytic characteristics for acidophytic and nitrophytic lichens	Completed	In Kathmandu University, Nepal
4	Measurement of tree characteristics	Tree height, DBH, crown cover, slope, bark furrow	Influences of tree characteristics and microclimatic conditions on the abundance of lichens	Completed	On-site in Nepal
5	ALPHA sampler deployment	Monitor atmospheric ammonia concentration	Annual average atmospheric ammonia concentrations to ground-truth modelled data, and to determine toxicity threshold levels of ammonia	Ongoing, till February 2027	UK Centre for Ecology & Hydrology
6	Biosocial survey	Document indigenous people and local communities' perception of lichens and atmospheric pollution	Rationale behind the importance of this project to be carried out in the Himalayan forest ecosystems and its implication for local communities.	Completed	On-site in Nepal
7	Deposition of duplicated herbarium specimens	Reference for collections at the National Herbarium of Nepal	Evidence for the collection and export of samples abroad	Completed	KATH, Nepal

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Team

Suman Prakash Pradhan (Lead), Sujan Prakash Pradhan (Field Support), Asmita Shrestha (Field Support), Swati Jha (Student Support), Shristi Kayastha (Student Support), Nani Raut (Local Mentor), Sanjeev Kumar Rai (Local Mentor), Christopher J Ellis (Supervisor), David S Stevenson (Supervisor), Ajinkya G Deshpande (Supervisor), Matthew R Jones (Advisor), Mark A Sutton (Supervisor) and Gothamie Weerakoon (Advisor).

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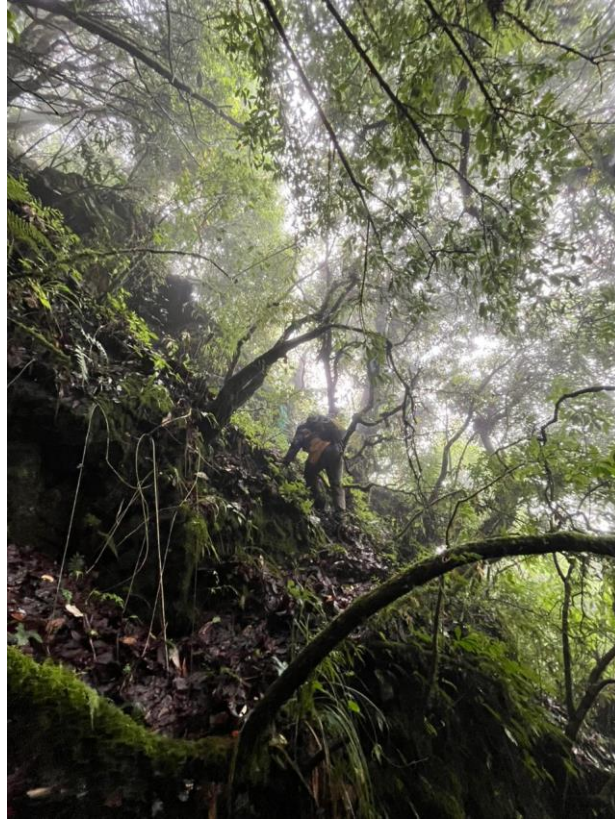
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Pictures



Field sites with complex topography and dense forest

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Struggle in the forest to get suitable trees for lichen sampling



Sampling of lichens

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Lichens on the trunk and the branch



Looking for tree characteristics

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Some macrolichens found during the field



Microlichens in lens

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Air-dried lichen specimens



Deploying ALPHA samplers for atmospheric ammonia monitoring

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Biosocial survey with indigenous people



Happy faces after the accomplishment of fieldwork