

**RELATIONSHIP OF TOPOGRAPHY AND SOIL CHEMISTRY
WITH SPECIES RICHNESS AND SPECIES COMPOSITION IN
CHANDRAGIRI HILL, CENTRAL NEPAL**



Submitted by

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DECLARATION

I hereby declare that the dissertation work entitled “**Relationship of topography and soil chemistry with species richness and species composition in Chandragiri hill, central Nepal**” is carried out by myself and has not been submitted elsewhere for any other academic degree. All the sources of information have been specifically acknowledged by reference wherever adopted from other sources.

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RECOMMENDATION LETTER

This is to recommend that the thesis entitled “**Relationship of topography and soil chemistry with species richness and species composition in Chandragiri hill, central Nepal**” has been carried out by Parvati Gharti for the partial fulfillment of degree of Master of Science in Botany. This is her original work and has been carried out under my supervision. To the best of my knowledge, this thesis work has not been submitted for any other degree in any institutions.

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LETTER OF APPROVAL

This thesis work submitted by Ms. Parvati Gharti, T.U. registration no. 5-2-22-2014 entitled “**Relationship of topography and soil chemistry with species richness and species composition in Chandragiri hill, central Nepal**” has been accepted as a partial fulfillment for the requirements of degree of Master of Science in Botany.

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LIST OF ABBREVIATIONS

AIC:	Akaike Information Criterion
ANOVA:	Analysis of Variance
AP:	Available Phosphorus
AK:	Available Potassium
CCA:	Canonical Correspondence Analysis
DCA:	Detrended Correspondence Analysis
GLM:	Generalized Linear Model
SOM:	Soil Organic Matter
SD:	Standard Deviation
SR:	Species Richness
TN:	Total Nitrogen

ABSTRACT

The present study aims at finding the effects of topography and soil chemistry with species richness and species composition in Chandragiri hill, central Nepal. Environmental variables like topography, soil, climate affect the distribution of species. Most of the literature reported a hump-shaped pattern between species richness and environmental variables. The study was carried out in the community forest of Chandragiri hill, central Nepal. Altogether, 35 plots of $10 \times 10 \text{ m}^2$ were established between 1500-2500 m. Quadrats of $10 \times 10 \text{ m}^2$, $5 \times 5 \text{ m}^2$ and $1 \times 1 \text{ m}^2$ were established for the assessment of trees, shrubs and herbs respectively. Soil was collected from the four corners of the plot and further analysis was carried out in the lab. Soil chemical properties i.e. pH, total nitrogen (TN), soil organic matter (SOM), available phosphorous (AP) and available potassium (AK) were determined in the laboratory. Altogether, 180 species were recorded from the study site. Data analysis was carried out using correlation, ANOVA, regression and ordination. Among the various variables studied, only soil pH showed a significant negative relationship with total species richness. Shrubs, herbs and climbers also showed the same pattern to soil pH as found between total species richness and soil pH. However, the tree showed no significant trend with soil pH. Herb and shrub showed significant relation with SOM and TN respectively. Species composition in the site was strongly influenced by altitude, pH, TN and AK. At the local level, soil heterogeneity was the factor determining species richness. On the other hand, species composition was influenced by the interaction of various environmental variables. Such studies provide information about species richness in the study site and hence can be precious for the formulation of conservation strategies. In addition, it also helps in prioritizing the site on the basis of biodiversity.

Keywords: diversity, pattern, environmental variables, composition, nutrients

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CHAPTER ONE

1. INTRODUCTION

1.1 Background

Evaluation of species richness aids to understand biodiversity as well as to formulate strategies for conservation in order to combat threats of biodiversity. The factors influencing species richness are of major concern in biodiversity and ecological research (Gaston 2000) due to increasing risk of extinction as a result of climate change and anthropogenic activities. Species richness is an indicator of biodiversity (Peet 1974). Environmental variables like topography, climate and soil are the factors determining species richness (Kharakwal et al. 2005). All the species are not adapted to the same environment. Different plants have different needs for light, temperature, moisture and nutrients and changes in these environmental variables cause diversity in habitats (Peringer et al. 2017) and hence strongly influence plant diversity and species composition (Mellado and Zamora 2015). Even small changes in environmental factors can have profound effects on species composition and diversity (Luan et al. 2012) however because of spatial and temporal variation in various environmental factors, it is challenging to assess the actual effect of these environmental factors (Kersti et al. 2020).

Climate is the critical factor determining species richness at both landscape and regional scale while plant species richness is governed by environmental heterogeneity at local scale (Lavers and Field 2006). Climate influences the availability of resources needed for plant growth such as moisture, temperature and light. On the other hand, heterogeneity in the environment may results in diversity in niche, allowing the co-existence of many species. Studies in tropical forests stressed moisture as a limiting factor in determining species richness (Gentry 1982) while studies from temperate regions emphasized on energy (Currie 1991).

To explain the relationship of species richness with the environment many hypotheses have been proposed. According to the water-energy hypothesis (Francis and Currie 2003), regions with higher availability of water and energy can favor more species

(Gaston 2000). The physiological tolerance hypothesis (Connell and Orias 1964) on the other hand states that species richness is greater in environments with suitable temperature conditions and water availability as more species can tolerate such conditions. Few species can survive in extremely hot or cold conditions. The mid-domain effect hypothesis states that species richness will increase from edges to mid-domain as more species ranges will overlap near the middle of the domain than at the edges. The species-area relationship proposes that with increase in the sampling area, more species will occur (Losos and Schluter 2000).

Topographic factors (altitude, slope and aspect) are often used in predicting species richness. These three factors together determine the microclimate and spatial distribution of species. Topographic variables affect species by controlling spatial distribution of solar radiation and precipitation (Iturrate-Garcia et al. 2016, Fan et al. 2020). The present study considered altitude, aspect and slope under topographic factors. Along the elevation gradient, many components of climate (such as temperature and rainfall) and local variables change and create variation in species richness (Lomolino 2001). Species need to adapt to the different climate along the elevation in order to survive. Due to the inability to adjust in different climatic conditions, the majority of the species are restricted to a particular elevation.

Aspect controls the species richness pattern by affecting the incidence of solar radiation. It is generally found that in the Northern Hemisphere southwest facing slopes have a warmer microclimate than northeast facing slopes (Perrings 1959) and vice versa on the Southern Hemisphere (Kutiel et al. 1998). The variation in the slope and aspect affects species richness (Boyko 1947, Carmel and Kadmon 1999, Nepali et al. 2021) by creating variation in the soil moisture, temperature, nutrients cycling and energy dissipation (Mohammad 2008). Aspect affects the availability of nutrients in soil by altering the rate of decomposition and affecting the soil microbial activity (Nahidan et al. 2015). The slope aspect results in the formation of distinct microclimatic conditions by influencing the spatial distribution of incoming solar radiation, temperature, precipitation, nutrient concentration (Gutiérrez-Jurado et al. 2006) associated with variation in soil properties. In studies conducted on east and west slopes, it has been reported solar radiation are tilted towards west, as a result west facing slopes are much more warmer than east-facing slopes (Bennie et al. 2006)

Slope also plays an important role in determining species richness through its influence on soil properties as soils on steep slopes are less moist and more acidic. With increase in steepness of slope, the soil becomes thinner and infertile as surface run-off washes topsoil easily (Liu et al. 2020).

Soil provides the medium for plant growth. Forest soil is a reservoir of nutrients for plants. Soil nutrients are essential for plant growth and development (de Jager et al. 2016) and also affect the diversity and species composition of plant communities (Becknell and Powers 2014). Plant species richness is affected by soil properties in numerous ways. Availability of nutrients results in less abiotic stress, hence allowing a number of species to coexist (Kepfer-Rojas et al. 2019). However, some studies reported that competitive species capable of efficiently utilizing resources are favored by high nutrient content (Dingaana et al. 2017). Decline in diversity occurs as a result of dominance of competitive species. Soil affects plant through two ways: direct and resource effects (Austin 2002, Pausas et al. 2003). Direct effect is related to pH whereas resource effect is through availability of nutrients and moisture.

Soil pH plays an essential role in affecting plant growth by influencing nutrients availability, nutrient toxicity and microbial activity. Most of the plants nutrients are available at slightly acidic to slightly alkaline soil pH (6.5 – 7.5). In both extremely acidic and alkaline soils, a number of nutrients become unavailable to plants due to the different reactions in soil which fix the nutrients. As a result of which species richness declines. Soil microbes through the mineralization process increase the availability of nutrients and survival of the soil microbial population is dependent on soil pH. Hence, soil pH is an important factor in determining nutrients availability to plants (Hajabbasi 1997).

Species richness is simply the number of species found in an area. On the other hand species diversity is the combination of both species richness and species abundance. Diversity indices help in knowing the cumulative effects of both richness and abundance. These include Shannon-Wiener diversity index (Shannon-Wiener 1963), Simpson diversity index (Simpson 1949) and Pielou evenness index (Pielou 1966). However, these indices have their own pros and cons.

Species belonging to the same life-form use similar resources and respond to the environment in a similar way. As a result, competition is expected to be less between

different life-forms as compared to within the same life-form. A comparison of different life-forms enables a finer and precise picture of causal factors than considering total species richness. Studies based on life-form for elevation and aspects have been done in the past but such studies considering soil factors were not available.

Most of the protected areas situated in Nepal are in lowland or highland physiographic regions. However, there are sites situated in mid-hills like Chandragiri hill that are important from a biodiversity point of view. It represents the true mid-hills of Nepal. Local people and indigenous people are dependent on forest resources for their livelihood. Due to tourism, human encroachment, construction works, a large portion of plant species are being destroyed. The information generated from the study can be valuable for forest managers to formulate strategies for the conservation and preservation of the forest.

1.2 Research questions

- Which environmental variable is the most important in governing species richness?
- How does species richness of different life-form vary with different environmental variables?

1.3 Hypothesis

- A hump-shaped pattern is expected between species richness and environmental variables.

1.4 Objectives

The general objective of the study was to evaluate the effect of topography and soil chemistry on species diversity. The specific objectives were

- To find out the factor best explaining species richness in the study area
- To evaluate the pattern of species richness based on life-form

1.5 Rationale of the study

The outcome of the study helps to understand the effect of the selected environmental variables i.e. topography and soil on species diversity. Such studies will assist in

knowing the status of the forest. While carrying out development works, using such information a decision can be made whether to perform the concerned work or not by prioritizing the biodiversity rich places.

Chandragiri hill located in the southwest of the Kathmandu valley is rich in biodiversity and is one of the famous hiking destinations in Nepal. It has been threatened and adversely affected due to tourism, development activities. So this study will provide information regarding the biodiversity of the forest in Chandragiri hill and will help concerned authority to formulate strategies necessary for the conservation of forest. Moreover, local people are dependent for their needs and survival on such forest resources so it is necessary to have an idea about the conditions of such forest. It will help in taking timely actions to avoid the degradation of forests.

Most of the works done in the past in Chandragiri hill were concerned with wildlife assessment (Katuwal et al. 2020), regeneration patterns (Dani and Baniya 2022) and carbon stock assessment (Gurung et al. 2022). Plant species richness assessment of the area has not been found in the previous literature and so the information regarding the species diversity of the area is lacking.

1.6 Limitation

- Due to differences in altitudinal gradient of different community forests, the number of plots sampled in different community forests were not uniform

CHAPTER TWO

2. LITERATURE REVIEW

2.1 Species richness and topography

Topography results in the formation of distinct micro-climatic conditions by influencing spatial distribution of light, heat, water and soil nutrients. In the present study three factors (altitude, slope and aspect) under topography were concerned.

2.1.1 Altitude

Rahbek (2005) identified three patterns of species richness (a monotonic decline with increasing elevation, a plateau at low elevation and a hump-shaped curve with peak at mid-elevation) after reviewing 204 studies conducted along elevation gradients. Many studies reported a hump-shaped pattern between species richness and altitude (Carpenter 2005, Das et al. 2020, Sharma et al. 2019, Song and Cao 2017). Bhattarai and Vetaas (2003) reported a hump-shaped pattern along altitude in sub-tropical elevation gradient of 100-1500 m in the Himalayas, east Nepal. The maximum species richness reported between 600-800 m. Kharkhwal et al. (2005) examined plant species richness between 200 and 5800 m in the Indian Central Himalaya. The peak richness was recorded between 1400-1600 m. Chawla et al. (2008) reported a maximum number of species in the range between 2501-3400 m in the study conducted in Bhabha valley, western India.

Nepali et al. (2021) studied the role of altitude on species richness in Arghakhanchi, west Nepal and found a unimodal pattern of species richness with altitude. In a study by Song and Cao (2017), species richness showed a unimodal response to altitude with a peak around 800 m in sub-tropical Eastern China. Sharma et al. (2019) evaluated plant species richness patterns along elevation gradients of 500-3300 m in the Eastern Himalaya. A hump-shaped pattern was reported for all life forms. The maximum tree species richness occurred between 1000-1500 m, maximum shrub species richness between 1200- 2000 m and maximum herb species richness between 1500-2500 m. Das et al. (2020) assessed the species richness patterns along the altitudinal gradient in western Himalaya, India. They found a peak at middle altitude (1500-3000 m). Few species can tolerate the full spectrum of environmental

conditions at gradient extremes (Sánchez-González and López-Mata 2005) which might be the reason for hump-shaped pattern.

Sharma et al. (2009) reported a decrease in tree species richness along the altitude 1850-2800 m in a moist temperate forest of Garhwal Himalaya. Saikia et al. (2017) explored the plant diversity pattern in Arunachal Pradesh, northeast India along elevation gradient 87-4161 m. They observed a decrease in species richness with an increase in elevation. Cirimwami et al. (2019) observed monotonic decline in species richness for woody life forms and increasing pattern for herbaceous life form along the altitude of 810-2760 m in East African mountain forest. Teshome et al. (2020) found a decrease in species richness with elevation in dry Afromontane forest of Ethiopia. Decreasing species richness at higher elevation may be due to eco-physiological pressures such as low temperature, low productivity and shorter growing season (Vetaas and Grytness 2002). Lower temperature results in the dominance of species that can tolerate severe conditions.

2.1.2 Slope

A negative correlation was found between tree species richness and slope in moist temperate forest of Garhwal Himalaya (Sharma et al. 2009). Teshome et al. (2020) also reported a negative correlation between species richness and altitude in dry Afromontane forest. Yang et al. (2021) investigated the relationship between plant community types and environmental factors in evergreen-deciduous broadleaved mixed forest in northwestern Hubei province, China. They also reported a negative relation between species richness and slope. However, Sánchez-González and López-Mata (2005) reported positive correlation of species richness with degree of slope in Sierra Nevada, Mexico. Slope has a negative effect on plant richness as high degree of slope results in immense water drainage, soil washing and subsequent decrement in soil fertility.

2.1.3 Aspect

In the northern hemisphere north-facing slopes having latitude around 30°-55° tend to receive less direct sunlight than the south-facing slopes (Searcy et al. 2003). As a result, the north aspect remains shaded for a longer period of time due to lower angle of sun. In mid-latitudinal region, the effect of aspect on vegetation is seen to be more pronounced. In temperate regions, aspect is the most influential factor. Heydari and

Mahdavi (2009) evaluated patterns of plant species diversity related to physiographic factors in Iran. They revealed south and southwest facing slopes had higher diversity in comparison to other aspects. This could be due to the increase in light level in the forest floor as a result of lower tree cover density in those aspects. Zeng et al. (2014) studied the effect of slope aspect and position on plant diversity and spatial distribution in the hilly region of Mount Taihang, North China. They confirmed south facing slopes exhibited higher shrub species richness while north facing slopes showed higher herb species richness. Shrubs are better adapted to drier conditions while herbs are adapted to wet, fertile and well drained conditions. Therefore, herbs are found to be widely distributed in northern slopes while shrubs in southern slopes.

Mahmoudi et al. (2018) investigated the impact of aspect on species diversity in western zone of Iran. Herbaceous species richness was found to be higher in the south than other three aspects. In comparison to lower latitude, at mid-latitude the effects of slope aspects on vegetation are seen to be pronounced. North-facing slopes are characterized with thick and dense vegetation whereas south-facing slopes are supported with scattered and thin vegetation (Singh 2018). Yang et al. (2020) investigated the effect of slope aspect on vegetation attributes in a mountainous dry valley in southwest China. They found north-facing slopes are associated with higher species diversity than south-facing slopes.

2.2 Species richness and soil chemical properties

2.2.1 Soil pH

Soil pH is one of the most important soil properties influencing plant growth. The pH of the soil affects plant growth through its effect on availability of nutrients for plants. Between pH 6-8, nitrogen availability is maximum because this pH range favors the soil microbes that decompose organic matter and organisms that fix nitrogen. It is noted that potassium, calcium and magnesium are widely available in alkaline soils. As a result of the decreasing cation exchange capacity and decreased amount of exchangeable nutrient cations, these nutrients become less available as acidity increases (Foth 1990). Species richness declines towards both acidic and alkaline soils. Not all the species are adapted to exploit highly acidic and alkaline soils and may require a narrow range of pH to survive (McFarland et al. 2015).

Vetaas (1997) reported that vascular plant richness was positively related to pH. Many studies found an increase in species richness with increase in pH (Roem and Berendse 2000, Stevens et al. 2004, Weiher et al. 2004) in grassland. Gould and Walker (1999) found a unimodal relationship between species richness and pH for vascular plants along a Canadian Arctic river. Some studies reported a hump-shaped pattern (Schuster and Diekmann 2003, Tyler 2003). Dingaana et al. (2017) investigated the relationship between soil chemical properties and plant diversity in South Africa. They observed a negative correlation between species richness and soil pH.

2.2.2 Soil nutrients

A hump-shaped pattern was reported between species richness and nutrient availability in many studies (Ashton 1977, Grime 1973, Huston 1980, Vermeer and Berendse 1983). At low nutrients, species richness is low, at intermediate level reaches to a peak and at high nutrient levels decline more gradually. Fewer species are able to tolerate extreme conditions of nutrient deficiency (Grime 1973). Species richness increases with resources as more species can survive. Few species become dominant and suppress others at high nutrient levels. Decline in species richness occurs due to competitive exclusion. Zhao et al. (2019) studied the relationship between soil nutrients and plant diversity in China. They confirmed a hump-shaped curve between species richness and soil nutrients.

Gairola et al. (2012) reported a negative correlation between species richness and K in moist temperate valley slopes of Garhwal Himalaya, India. Nadeau and Sullivan (2015) studied the relationship between plant biodiversity and soil chemical fertility in a mature tropical forest of Costa Rica. They found a negative correlation between richness and soil potassium, phosphorous, calcium contents. Huston (1980) also reported a decrease in tree species richness in tropical forest. In general, less fertile soils favors high tree species richness. Due to lack of resources in low fertility soil, strong competitors become unable to outcompete others hence, resulting in higher species richness. Yang et al. (2021) investigated the relationship between plant community types and environmental factors in evergreen-deciduous broadleaved mixed forest in northwestern Hubei province, China and also reported a negative relation between species richness with available phosphorous and total nitrogen. With increasing nutrient availability, plant diversity is known to decline (Critchley et al.

2002a, Cornwell and Grubb 2003). A small number of competitive species capable of utilizing resources rapidly and accumulating biomass are favored by high nutrient availability in grassland (Critchley et al. 2002b). Similarly many researches showed decrease in species richness after soil fertilization (Mittelbach et al. 2001).

Janssens et al. (1998) while evaluating the relationship between richness and soil chemical properties in temperate grassland reported a positive relationship between richness and extractable P and K in soil. Kumar et al. (2010) explored the relationship between tree species richness and soil nutrient concentration in three different sites in a dry deciduous forest in western India. A positive correlation between tree species richness and concentrations of nitrogen, phosphorous and carbon was reported. A positive relation between tree species richness and calcium, phosphorous and potassium contents is confirmed by many studies in temperate forests. Bulenga et al. (2021) studied the relationship between tree species diversity with soil chemical properties in semi-dry Miombo woodland ecosystems of Tanzania and found species richness was positively correlated with total nitrogen and available phosphorous. Soil organic matter provides the nutrients to plants. Through the decomposition process nutrients in soil organic matter are made available to plants. Soil organic matter affects species composition by having an important influence on soil physical and chemical properties, soil fertility status, plant nutrition and biological activity in the soil (Syaed 2021). Phosphorous is required by plants in large amounts since its concentration and availability determines the soil fertility and productivity (Yang et al. 2021).

CHAPTER THREE

3. MATERIALS AND METHODS

3.1 Study area

The study was carried out in five adjoining community forests of Chandragiri hill. It comprises an area of about 11 km² in Chandragiri Municipality, Kathmandu. It is about 16 km from the central Kathmandu. It lies between 27° 38' N to 27° 43' N latitude and 85° 11' E to 85° 16' E longitude (Figure 1). The elevation ranges from 1300 to 2540 m above sea level.

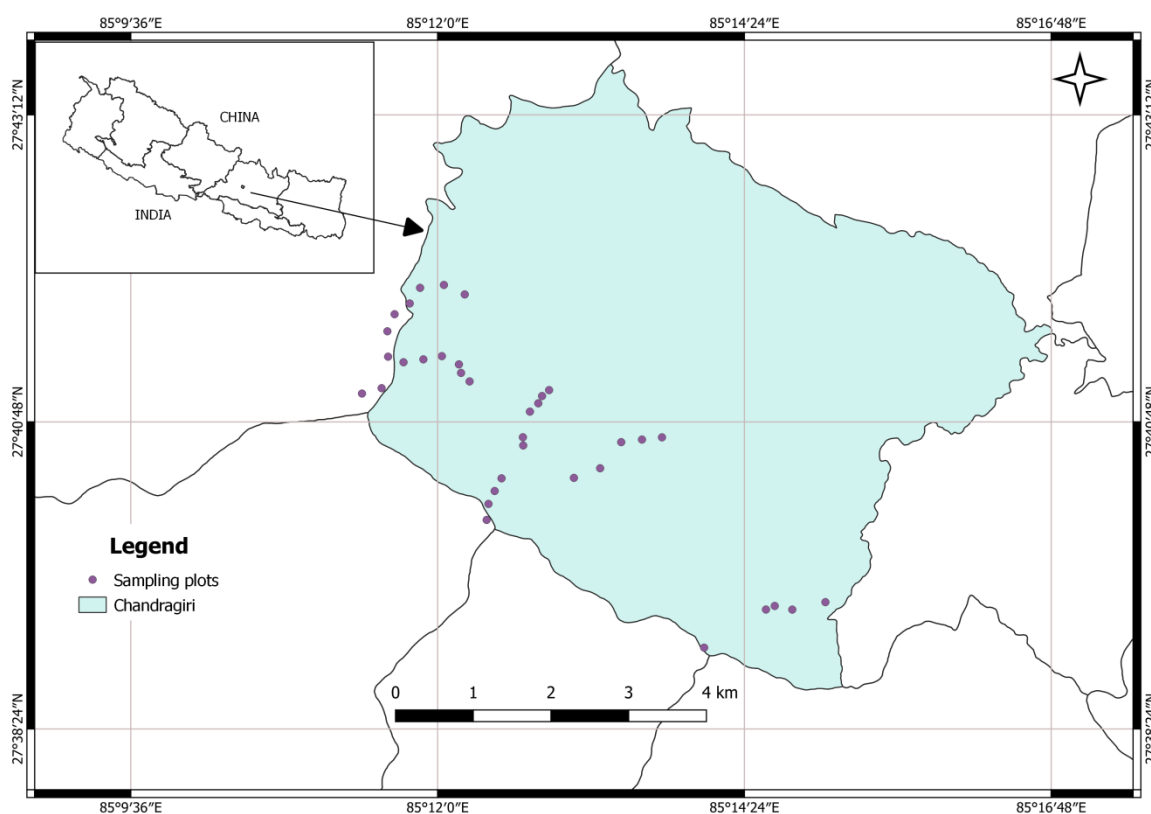


Figure 1: Map of study area (circles are showing the plots sampled)

The climate of the study area is subtropical to temperate type. The warm season lasts from April to October and the cold season from December to February. The maximum and minimum average monthly temperature was reported to be in the months of June (28.9 °C) and January (18.3 °C) respectively. The highest (341.8 mm) average monthly rainfall was reported to occur in the month of July whereas the

lowest (5.5 mm) average monthly rainfall occurred in the month of November (Figure 2).

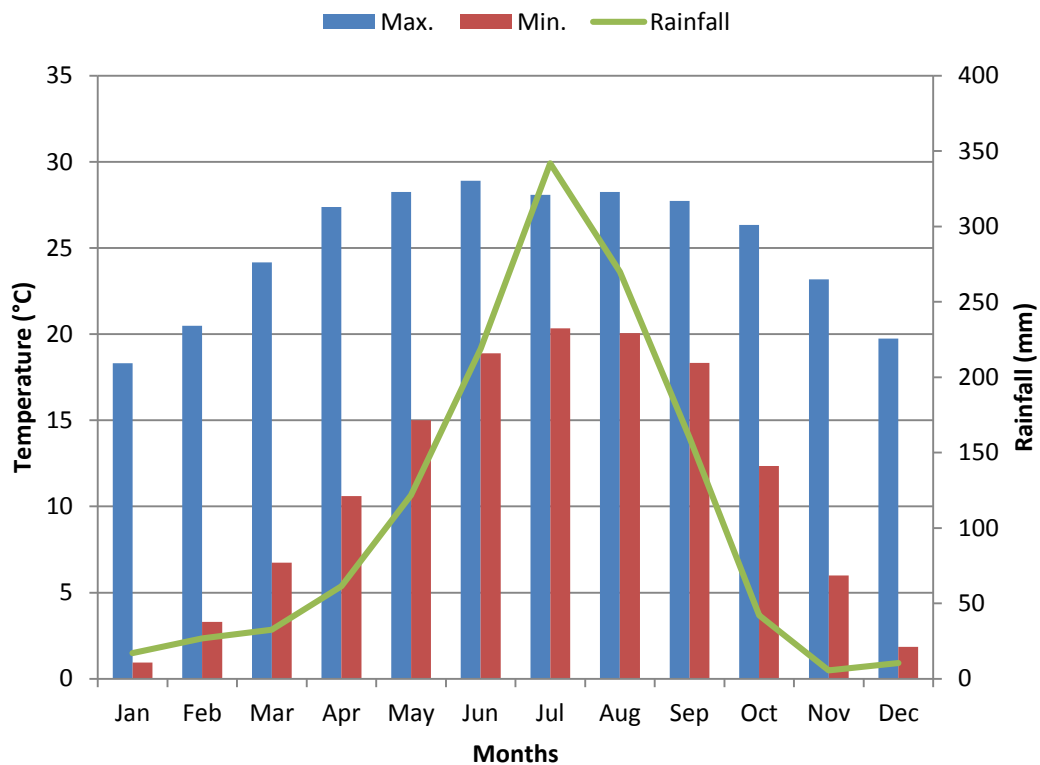


Figure 2: Thirty years (1993-2022) average monthly temperature and rainfall recorded at Khokana weather station. (Source: Department of Hydrology and Meteorology, Babarmahal, Kathmandu)

The study area is dominated by broad-leaved mixed forest. At the lower elevation, *Alnus nepalensis* is a dominant species. Species like *Albizia* sp., *Ligustrum confusum*, *Schima wallichii*, *Maesa chisia*, *Pinus roxburghii* etc were well distributed in this part. At higher elevation, species like *Rhododendron arboreum*, *Quercus lanata* and *Quercus semecarpifolia* were dominant.

3.2 Field sampling

The field work was carried out in 35 plots during the winter season from February to March in 2022. The vegetation sampling was carried out using quadrat method (Cottam and Curtis 1956). The plots were established at vertical 100 m intervals. At each site a quadrat of $10 \times 10 \text{ m}^2$ for trees, $5 \times 5 \text{ m}^2$ for shrubs and $1 \times 1 \text{ m}^2$ for herbs was established (Nanda et al. 2018). Within each quadrat of $10 \times 10 \text{ m}^2$, four quadrats

($5 \times 5 \text{ m}^2$) for shrubs and four quadrats ($1 \times 1 \text{ m}^2$) for herbs were selected. Hence, the field survey included 35 quadrats of $10 \times 10 \text{ m}^2$ for trees, 140 quadrats of $5 \times 5 \text{ m}^2$ for shrubs and 140 quadrats of $1 \times 1 \text{ m}^2$ for herbs.

Environmental variables such as altitude, latitude, longitude, aspect and slope were noted for each plot. Altitude, aspect, latitude and longitude were measured using GPS and the slope was determined using a clinometer. Within each plot associated trees, herbs, shrubs and climbers were identified. The number of individuals of each species were counted and noted down.

Most of the plants were identified in the field and those that were not identified; their local names were asked from the local people to make the task of plant identification easier. Specimens of all unidentified plants sampled in the plot were collected, tagged and later on pressed using herbarium press. Unidentified species were identified with the help of existing literatures (DPR 1986, Press et al. 2000, Shrestha et al. 2018), respective local names and taxonomic experts.

The soil samples were collected from the four corners of the plot at a depth of 0-10 cm and mixed to prepare one composite soil sample. The plant materials and pebbles in each sample were separated by hand and removed. Approximately, 500 gm of soil sample was collected from each location and placed into plastic bags.

3.3 Laboratory analysis

The collected soil samples were transferred to the laboratory for further analysis. The collected soil samples were air dried for two weeks and passed through a 2 mm sieve prior to the laboratory analysis. Soil pH using a pH meter with a soil to water ratio of 1:2.5 (Pradhan 1996), soil organic matter by Walkley and Black method (Walkley and Black 1934), total nitrogen by Kjeldahl method (Bremner 1960), soil phosphorus by Bray and Kurtz method (Bray and Kurtz 1945) and soil potassium by flame photometer (Jackson 1967) were measured.

3.3.1 Soil pH

Ten grams of air dried soil samples were weighed and kept in a 50 ml beaker and 25 ml of distilled water was added. The beaker was shaken for one minute and left for an hour. The pH meter was calibrated using standard buffer solutions of pH 4.0 and 7.0. The pH of soil suspension was then measured with the calibrated pH meter.

3.3.2 Soil total nitrogen

Digestion: One gram of air dry and sieved soil was weighed in a clean and dry kjeldahl flask. 3.5 gram potassium sulfate and 0.4 gram of copper sulfate were weighed and the mixture was then transferred to the kjeldahl flask containing soil. Six ml of concentrated sulphuric acid was added in the soil mixture with gentle shaking. The flask was placed on the pre-heated mantle. After 30 minutes, the color of the mixture was checked. After the appearance of green color the flask was removed from the mantle and allowed to cool down. 50 ml of distilled water was added to the flask after 15-20 minutes of cooling and then the mixture was shaken.

With each batch of digestion, a single blank (without soil) was included.

Distillation: The digest was transferred to kjeldahl distillation flask. Ten ml of boric acid indicator was taken in a clean and dry beaker (100 ml) and placed below the nozzle of the condenser in such a way that the end of the nozzle dipped into the indicator. Once the digest became warm, 30 ml of sodium hydroxide was added. Once the distillate began to condense, the color of the boric acid indicator changed from pink to green. The distillation was continued until the volume of distillate in the beaker reached to about 50 ml.

Titration: The beaker containing distillate was removed and titrated with hydrochloric acid.

$$\text{Nitrogen in soil (\%)} = \frac{14 \times N \times (S - B) \times 100}{\text{Weight of soil (mg)}}$$

Where, N= normality of HCl

S= volume of HCl consumed with sample (ml)

B= volume of HCl consumed with blank (ml)

3.3.3 Soil organic matter

0.25 gram of soil was taken in a 500 ml conical flask. Five ml of potassium dichromate was pipetted in and swirled a little. Then, ten ml of sulphuric acid was added and swirled again two to three times. The flask was allowed to stand for 30 minutes and then 100 ml of distilled water was added. 5 ml of phosphoric acid and 1ml of diphenylamine indicator were added. The contents were titrated with 0.5 N

ferrous ammonium sulfate solution till the color changed from blue-violet to green. Simultaneously, a blank was run without soil.

$$\text{Carbon in soil (\%)} = N \frac{(\text{Blank reading} - \text{Soil reading})}{\text{Weight of soil (gm)}} \times 0.003 \times 100$$

Where, N = normality of ferrous ammonium sulfate

$$\text{Organic carbon} = \text{organic carbon estimated} \times 1.3$$

$$\text{SOM} = \text{organic carbon} \times 1.724$$

3.3.4 Soil available phosphorus

Five grams of air dried soil sample was taken in a conical flask and 50 ml of Bray extracting solution was added to it. The suspension was shaken for five minutes and then was filtered. Five ml of the aliquot of the extract was taken in a 25 ml volumetric flask. The distilled water to 20 ml and then four ml Murphy Riley solution were added. After 15 minutes, the intensity of blue color was read using 730 nm on a spectrophotometer.

$$\text{Phosphorous in soil (ppm)} = \frac{\text{graph (ppm)} \times \text{volume of extractant} \times \text{volume made} \times 2}{\text{Weight of soil} \times \text{aliquot taken}}$$

3.3.5 Soil available potassium

Two grams of soil was shaken with 20 ml neutral normal ammonium acetate for five minutes. The soil solution was immediately filtered through filter paper. A standard curve of potassium was prepared by aspirating 0, 5, 10, 15, 20 and 25 ppm after adjusting the full scale deflection of the flame photometer with 25 ppm potassium. The readings were noted and a graph was drawn. The soil solution was aspired, its reading was noted and potassium in the soil solution was determined from the graph.

3.4. Data analysis

A data matrix consisting of eight environmental variables for each 35 plots and species data matrix consisting of 180 species by 35 plots was prepared for data analysis using Microsoft Excel. The topographic variables included in the study were elevation, slope and aspect whereas the soil variables were pH, total nitrogen (TN), soil organic matter (SOM), available phosphorus (AP) and available potassium (AK).

The species richness, Shannon Wiener diversity index, evenness and Simpson index were calculated using the ‘vegan’ R package (Oksanen et al. 2007). Species richness is simply taken as the number of species present in each plot sampled. However, species diversity takes into account both species richness and species evenness. Evenness refers to the uniformity in the number of individuals within species. Diversity indexes were calculated to evaluate the relationship of species richness with different diversity indices.

Margalef index is similar to species richness but also takes into account the number of individuals present within the species. It was calculated using the formula (Margalef 1958)

$$M = \frac{S-1}{\ln N}$$

Where S = number of species and N = number of individuals

Shannon-Weiner diversity index is the most commonly used diversity index in ecology. It takes into account both species richness and evenness. It assumes all species are represented in a sample and that they are randomly sampled. Its value usually ranges from 1.5 to 3.5. The Shannon Wiener diversity index (Shannon & Wiener 1963) was calculated using the formula

$$H = \sum_{i=1}^s p_i \ln p_i$$

Where p_i is the proportion of species

Evenness indicates how homogeneous a site is in terms of the abundance of its species. Its value varies from zero to one where value one represents the condition of all species being equally abundant and zero indicates no evenness. The Pielou evenness index (Pielou 1966) was calculated by using the formula

$$E = H / \ln S$$

Where H = Shannon Wiener diversity index and S = total number of species

Simpson index is a dominance index since it is biased towards common species and less sensitive to rare species. Simpson diversity index is calculated by subtracting Simpson index from one. It measures the probability of two individuals selected

randomly belonging to different species. Simpson diversity index (Simpson 1949) was calculated using the formula

$$D = 1 - \sum_{i=1}^s p_i^2$$

Where p_i is the proportion of species

To examine the normality of the response variable, a Shapiro-Wilk test was performed. One way analysis of variance was used to find the significant difference in species richness with different environmental variables. Pearson's correlation was used to evaluate the correlation between species richness and environment variables.

Generalized linear model (McCullagh and Nelder 1989) was used to estimate the relationship between species richness and different environmental variables. To deal with overdispersion of the deviance, quasi poisson distribution was used. Model selection was done using Akaike Information Criterion (AIC) in a stepwise algorithm. The model was tested against the null model as well as up to the second order polynomial function. Using randomForest package (Liaw and Wiener 2002), mean decrease accuracy and importance value of different environmental variables were determined.

Multivariate analysis was used to evaluate the influence of topographic and soil factors on species composition. Detrended Correspondence Analysis (DCA) (Hill and Gauch 1980) of species data was first performed to determine whether a linear or unimodal model to be used. Since the gradient length of the first axis was greater than 2.5 standard deviation, a unimodal Canonical Correspondence Analysis (CCA) (ter Braak 1986) was used to evaluate the influence of environmental variables on species composition. Effects of environmental variables such as altitude, aspect, slope, pH, TN, SOM, AP and AK were investigated. All the analyses were performed in R software version 4.2.1 (R Core Team 2022).

CHAPTER FOUR

4. RESULTS

4.1 Summary of the variables

Altogether, there were eight environmental variables (three topographic and five soil) studied during the study (Table 1). Altitude in the site ranged from 1527 m asl to 2440 m asl. Likewise, the minimum slope of the plot sampled was 10° to the maximum slope of the plot sampled was 50°. The physical characteristics of the plots sampled are shown in Appendix I.

Table 1: Summary of the variables

Variables	Minimum	Maximum	Median	Mean	SD
Altitude (m)	1527	2440	1839	1900	266.92
Aspect (°)	4	359	68	94.43	96.34
Slope (°)	10.0	50.0	27.0	26.4	10.43
SR	12.0	45.0	23.0	24.1	7.96
pH	4.28	6.25	4.97	5.07	0.47
TN (g/kg)	0.70	6.40	2.80	2.87	1.34
SOM (g/kg)	18.80	126.40	80.06	80.81	31.26
AP (mg/kg)	1.63	10.13	4.59	4.86	2.38
AK(mg/kg)	19.0	208.0	104.0	110.0	53.53

4.2 Species diversity

A total of 180 species belonging to 142 genera and 68 families were documented from the study area (Table 2). The total species richness ranged from 12 to 45 with highest value being recorded in mid elevation (2051 m asl) and lowest in lower elevation (1829 m asl).

Table 2: List of species found in study area

S.N.	Botanical name	Family	Abbreviation
1.	<i>Achyranthes aspera</i> L.	Amaranthaceae	Achy aspe
2.	<i>Adiantum capillus-veneris</i> L.	Pteridaceae	Adia capi
3.	<i>Ageratina adenophora</i> (Spreng.) R. King & H. Rob.	Asteraceae	Ager aden
4.	<i>Ainsliaea latifolia</i> (D. Don) Sch. Bip.	Asteraceae	Ains lati
5.	<i>Albizia</i> sp	Fabaceae	Albi sp
6.	<i>Alnus nepalensis</i> D. Don	Betulaceae	Alnu nepa
7.	<i>Anaphalis contorta</i> (D. Don) Hook. fil.	Asteraceae	Anap cont
8.	<i>Anaphalis margaritacea</i> (L.) Benth.	Asteraceae	Anap maga
9.	<i>Apios carnea</i> (Wall.) Benth. ex Baker	Fabaceae	Apio carn
10.	<i>Aristolochia serpentaria</i> L.	Aristolochiaceae	Aris serp
11.	<i>Artemisia indica</i> Willd.	Asteraceae	Arte indi
12.	<i>Arundinella nepalensis</i> Trin.	Poaceae	Arun nepa
13.	<i>Asparagus setaceus</i> (Kunth) Jessop	Liliaceae	Aspa seta
14.	<i>Athyrium filix-femina</i> (L.) Roth	Athyriaceae	Athy fili
15.	<i>Berberis aristata</i> DC.	Berberidaceae	Berb aris
16.	<i>Berberis wallichiana</i> DC.	Berberidaceae	Berb wall
17.	<i>Bidens pilosa</i> L.	Asteraceae	Bide pilo
18.	<i>Boehmeria nivea</i> (L.) Gaudich.	Urticaceae	Boeh nive
19.	<i>Boehmeria platyphylla</i> Buch.-Ham. ex D. Don	Urticaceae	Boeh plat
20.	<i>Boenninghausenia albiflora</i> (Hook.) Rchb. ex Meisn.	Rutaceae	Boen albi

21.	<i>Buddleja asiatica</i> Lour.	Scrophulariaceae	Budd asia
22.	<i>Camellia kissi</i> Wall.	Theaceae	Came kiss
23.	<i>Capillipedium assimile</i> (Steud.) A. Camus	Poaceae	Capi assi
24.	<i>Carex baccans</i> Nees	Cyperaceae	Care back
25.	<i>Carex pendula</i> Huds.	Cyperceae	Care pend
26.	<i>Carex</i> sp.	Cypeaceae	Care sp
27.	<i>Castanopsis indica</i> (Roxb. ex Lindl.) A. DC.	Fagaceae	Cast indi
28.	<i>Castanopsis tribuloides</i> (Sm.) A. DC.	Fagaceae	Cast trib
29.	<i>Celtis australis</i> L.	Cannabaceae	Celt aust
30.	<i>Cheilanthes farinosa</i> (Forssk.) Kaulf	Pteridaceae	Chei fari
31.	<i>Chromolaena odorata</i> (L.) R. King & H. Rob.	Asteraceae	Chro odor
32.	<i>Cinnamomum tamala</i> (Buch.-Ham.) T. Nees & Eberm.	Lauraceae	Cinn tama
33.	<i>Clematis connata</i> DC.	Ranunculaceae	Clem conn
34.	<i>Clematis vitalba</i> L.	Ranunculaceae	Clem vita
35.	<i>Cleyera japonica</i> Thunb.	Theaceae	Cley japo
36.	<i>Codariocalyx motorius</i> (Houtt.) H. Ohashi	Fabaceae	Coda moto
37.	<i>Coniogramme intermedia</i> Hieron.	Pteridaceae	Coni inte
38.	<i>Coriaria nepalensis</i> Wall.	Coriariaceae	Cori nepa
39.	<i>Cornus oblonga</i> Wall.	Cornaceae	Corn oblo
40.	<i>Crassocephalum crepidioides</i> (Benth.) S Moore	Asteraceae	Cras crep
41.	<i>Crotalaria cytisoides</i> Roxb. ex DC.	Fabaceae	Crot cyti
42.	<i>Daphne bholua</i> Buch.-Ham. ex D. Don	Thymelaeaceae	Daph bhol
43.	<i>Desmodium elegans</i> DC.	Fabaceae	Desm eleg

44.	<i>Desmodium uncinatum</i> (Jacq.) DC.	Fabaceae	Desm unci
45.	<i>Dicranopteris linearis</i> (Burm. fil.) Underw.	Gleicheniaceae	Dicr line
46.	<i>Dioscorea deltoidea</i> Wall. ex Griseb.	Dioscoreaceae	Dios delt
47.	<i>Dodecadenia grandiflora</i> Nees	Lauraceae	Dode gran
48.	<i>Drepanostachyum falcatum</i> (Nees) Keng f.	Poaceae	Drep falc
49.	<i>Elatostema lineolatum</i> Wight	Urticaceae	Elat line
50.	<i>Eragrostis tenella</i> (Linn.) P. Beauv. ex Roem. & Schult.	Poaceae	Erag tene
51.	<i>Eriobotrya dubia</i> (Lindl.) Decne.	Rosaceae	Erio dubi
52.	<i>Eurya acuminata</i> DC.	Theaceae	Eury acum
53.	<i>Eurya cerasifolia</i> (D. Don) Kobuski	Theaceae	Eury cera
54.	<i>Ficus ottoniifolia</i> (Miq.) Miq.	Moraceae	Ficu otto
55.	<i>Ficus virens</i> Aiton	Moraceae	Ficu vire
56.	<i>Fragaria vesca</i> L.	Rosaceae	Frag vesc
57.	<i>Fraxinus angustifolia</i> Vahl	Oleaceae	Frax angu
58.	<i>Fraxinus floribunda</i> Wall.	Oleaceae	Frax flor
59.	<i>Galium mollugo</i> L.	Rubiaceae	Gali moll
60.	<i>Gaultheria fragrantissima</i> Wall.	Ericaceae	Gaul frag
61.	<i>Gentiana capitata</i> Buch.-Ham. ex D. Don	Gentianaceae	Gent capi
62.	<i>Geranium nepalense</i> Sweet	Geraniaceae	Gera nepa
63.	<i>Gerbera maxima</i> (D. Don) Beauv.	Asteraceae	Gerb maxi
64.	<i>Girardinia diversifolia</i> (Link) Friis	Urticaceae	Gira dive
65.	<i>Hedera nepalensis</i> K. Koch	Araliaceae	Hede nepa
66.	<i>Hedychium</i> sp.	Zingiberaceae	Hedy sp
67.	<i>Hedyotis scandens</i> Roxb. ex D. Don	Rubiaceae	Hedy scan

68.	<i>Holboellia latifolia</i> Wall.	Lardizabalaceae	Holb lati
69.	<i>Homalium napaulense</i> (DC.) Benth.	Flacourtiaceae	Homa napa
70.	<i>Hoya lanceolata</i> Wall. ex D. Don	Asclepiadaceae	Hoya lanc
71.	<i>Hydrangea febrifuga</i> (Lour.) Y. De Smet & C. Granados	Hydrangeaceae	Hydr febr
72.	<i>Hydrocotyle sibthorpioides</i> Lam.	Apiaceae	Hydr sibt
73.	<i>Hypericum</i> sp.	Clusiaceae	Hype sp
74.	<i>Ilex aquifolium</i> L.	Aquifoliaceae	Ilex aqu
75.	<i>Indigofera dosua</i> Buch.-Ham. ex D. Don	Fabaceae	Indi dosu
76.	<i>Inula cappa</i> (Buch.-Ham. ex D. Don) DC.	Asteraceae	Inul capp
77.	<i>Jasminum officinale</i> L.	Oleaceae	Jasm offi
78.	<i>Juglans regia</i> L.	Juglandaceae	Jugl regi
79.	<i>Koenigia mollis</i> (D. Don) T. M. Schust. & Reveal	Polygonaceae	Koen moll
80.	<i>Lantana</i> sp.	Verbanaceae	Lant sp
81.	<i>Lamium album</i> L.	Lamiaceae	Lami albu
82.	<i>Ligustrum sinense</i> Lour.	Oleaceae	Ligu sine
83.	<i>Lindenbergia grandiflora</i> (Buch.-Ham. ex D. Don) Benth	Scrophulariaceae	Lind gran
84.	<i>Lindera neesiana</i> (Wall. ex Nees) Kurz	Lauraceae	Lind nees
85.	<i>Lithocarpus elegans</i> (Blume) Hatus. ex Soepadmo	Fagaceae	Lith eleg
86.	<i>Lyonia ovalifolia</i> (Wall.) Drude	Ericaceae	Lyon oval
87.	<i>Machilus odoratissima</i> Nees	Lauraceae	Mach odor
88.	<i>Madhuca longifolia</i> (J. Koenig ex L.) J. F. Macbr.	Sapotaceae	Madh long

89.	<i>Maesa chisia</i> Buch.-Ham. ex D. Don	Myrsinaceae	Maes chis
90.	<i>Mahonia napaulensis</i> DC.	Berberidaceae	Maho napa
91.	<i>Mallotus philippensis</i> (Lam.) Mull.Arg	Euphorbiaceae	Mall phil
92.	<i>Maytenus</i> sp.	Celastraceae	Mayt sp
93.	<i>Melastoma malabathricum</i> L.	Melastomaceae	Mela mala
94.	<i>Molineria capitulata</i> (Lour.) Herb.	Hypoxidaceae	Moli capi
95.	<i>Mussaenda macrophylla</i> Wall.	Rubiaceae	Muss macr
96.	<i>Myrica esculenta</i> Buch.-Ham. ex D. Don	Myricaceae	Myri escu
97.	<i>Myrsine capitellata</i> Wall.	Myrsinaceae	Myrs capi
98.	<i>Myrsine seguinii</i> H. Lév.	Myrsinaceae	Myrs segu
99.	<i>Myrsine semiserrata</i> Wall.	Myrsinaceae	Myrs semi
100.	<i>Neocinnamomum caudatum</i> (Nees) Merr.	Lauraceae	Neoc caud
101.	<i>Neolitsea sericea</i> (Bl.) Koidz	Lauraceae	Neol seri
102.	<i>Onychium japonicum</i> (Thunb.) Kunze	Pteridaceae	Onyc japo
103.	<i>Onychium siliculosum</i> (Desv.) C. Chr.	Pteridaceae	Onyc sili
104.	<i>Ophiopogon</i> sp.	Convallariaceae	Ophi sp
105.	<i>Oplismenus burmanni</i> (Retz.) P. Beauv.	Poaceae	Opli burm
106.	<i>Osbeckia nepalensis</i> Hook.	Melastomaceae	Osbe nepa
107.	<i>Osyris wightiana</i> Wall. ex Wight	Santalaceae	Osyw wigh
108.	<i>Oxyspora paniculata</i> (D. Don) DC.	Melastomaceae	Oxys pani
109.	<i>Parthenium hysterophorus</i> L.	Asteraceae	Part hyst
110.	<i>Persea duthiei</i> King ex Hook. F.	Lauraceae	Pers duth
111.	<i>Persicaria chinensis</i> (L.) Nakai	Polygonaceae	Pers chin
112.	<i>Phyllanthus niruri</i> L.	Euphorbiaceae	Phyl niru
113.	<i>Phyllanthus reticulatus</i> Pair	Euphorbiaceae	Phyl reti

114.	<i>Pilea scripta</i> (Buch.-Ham. ex D. Don) Wedd.	Urticaceae	Pile scri
115.	<i>Pinus roxburghii</i> Sarg.	Pinaceae	Pinu roxb
116.	<i>Pinus wallichiana</i> A.B. Jacks.	Pinaceae	Pinu wall
117.	<i>Piper betle</i> L.	Piperaceae	Pipe belt
118.	<i>Piptanthus nepalensis</i> (Hook.) Sweet	Fabaceae	Pipt nepa
119.	<i>Polystichum polyblepharum</i> (Roem. ex Kunze) C. Presl	Dryopteridaceae	Poly poly
120.	<i>Polystichum squarrosum</i> (D. Don) Fée	Dryopteridaceae	Poly squa
121.	<i>Primula denticulata</i> Sm.	Primulaceae	Prim dent
122.	<i>Prunus caroliniana</i> (Mill). Aiton	Rosaceae	Prun caro
123.	<i>Prunus cerasoides</i> D. Don	Rosaceae	Prun cera
124.	<i>Prunus</i> sp.	Rosaceae	Prun sp
125.	<i>Prunus spinosa</i> L.	Rosaceae	Prun spin
126.	<i>Pteridium aquilinum</i> (L.) Kuhn	Pteridaceae	Pter aqu
127.	<i>Pteris latipinna</i> Y. S. Chao & W. L. Chiou	Pteridaceae	Pter lati
128.	<i>Pteris vittata</i> L.	Pteridaceae	Pter vitt
129.	<i>Pueraria peduncularis</i> Grah.	Fabaceae	Puer pedu
130.	<i>Pyracantha crenulata</i> (D. Don) M. Roem	Rosaceae	Pyra cren
131.	<i>Pyrus pashia</i> Buch.-Ham. ex D. Don	Rosaceae	Pyru pash
132.	<i>Quercus floribunda</i> Lindl. ex A.Camus	Fagaceae	Quer flor
133.	<i>Quercus glauca</i> Thunb.	Fagaceae	Quer glau
134.	<i>Quercus lamellosa</i> Sm.	Fagaceae	Quer lame
135.	<i>Quercus semecarpifolia</i> Sm.	Fagaceae	Quer seme
136.	<i>Quercus lanata</i> Sm.	Fagaceae	Quer lana
137.	<i>Randia tetrasperma</i> (Roxb.) Benth. & Hook.	Rubiaceae	Rand tetr

f. Brandis

138.	<i>Rhododendron arboreum</i> Sm.	Ericaceae	Rhod arbo
139.	<i>Rhus chinensis</i> Mill.	Anacardiaceae	Rhus chin
140.	<i>Rhus javanica</i> L.	Anacardiaceae	Rhus java
141.	<i>Rhus wallichii</i> Hook f.	Anacardiaceae	Rhus wall
142.	<i>Rosa sempervirens</i> L.	Rosaceae	Rosa semp
143.	<i>Roscoea purpurea</i> Sm.	Zingiberaceae	Rocs purp
144.	<i>Rubia manjith</i> Roxb.	Rubiaceae	Rubi manj
145.	<i>Rubus acuminatis</i> Sm.	Rosaceae	Rubu acum
146.	<i>Rubus ellipticus</i> Sm.	Rosaceae	Rubu elli
147.	<i>Rubus paniculatus</i> Sm.	Rosaceae	Rubu pani
148.	<i>Rubus ulmifolius</i> Schott	Rosaceae	Rubu ulmi
149.	<i>Sageretia thea</i> (Osbeck) Johnst.	Rhamnaceae	Saga thea
150.	<i>Sarcococca hookeriana</i> Baill.	Buxaceae	Sarc hook
151.	<i>Saurauia napaulensis</i> DC.	Actinidiaceae	Saur napa
152.	<i>Schima wallichii</i> (DC.) Korth.	Theaceae	Schi wall
153.	<i>Senecio wallichii</i> DC.	Asteraceae	Sene wall
154.	<i>Senecio scandens</i> (Buch.-Ham.)	Asteraceae	Sene scan
155.	<i>Smilax aspera</i> L.	Liliaceae	Smil aspe
156.	<i>Smilax elegans</i> Wall. ex Kunth	Liliaceae	Smil eleg
157.	<i>Smilax ferox</i> Wall. ex Kunth	Liliaceae	Smil fero
158.	<i>Smilax glauca</i> Walter	Liliaceae	Smil glau
159.	<i>Smilax lanceifolia</i> Roxb.	Liliaceae	Smil lanc
160.	<i>Smilax ovalifolia</i> Roxb.	Liliaceae	Smil oval
161.	<i>Smilax perfoliata</i> Lour.	Liliaceae	Smil perf

162.	<i>Smilax zeylanica</i> L.	Liliaceae	Smil zeyl
163.	<i>Solanum virginianum</i> L.	Solanaceae	Sola virg
164.	<i>Sophora glauca</i> DC.	Fabaceae	Soph glau
165.	<i>Stachys bullata</i> Benth.	Lamiaceae	Stac bull
166.	<i>Stauntonia</i> sp.	Lardizabalaceae	Stau sp
167.	<i>Stephania elegans</i> Hook. f. & Thomson	Menispermaceae	Step eleg
168.	<i>Strobilanthes wallichii</i> Nees	Acanthaceae	Stro wall
169.	<i>Synotis cappa</i> (Buch.-Ham. ex D. Don) C. Jeffrey & Y. L. Chen	Asteraceae	Syno capp
170.	<i>Syzygium cumini</i> (L.) Skeels	Myrtaceae	Syzy cumi
171.	<i>Tetrastigma bracteolatum</i> (Wall.) Planch.	Vitaceae	Tetr brac
172.	<i>Parthenocissus quinquefolia</i> (L.) Planch.	Vitaceae	Part quin
173.	<i>Thalictrum foliolosum</i> DC.	Ranunculaceae	Thal foli
174.	<i>Toddalia asiatica</i> Lam.	Rutaceae	Todd asia
175.	<i>Trachelospermum</i> sp.	Apocynaceae	Trac sp
176.	<i>Urtica dioica</i> L.	Urticaceae	Urti dioi
177.	<i>Viburnum cylindricum</i> Buch.-Ham. ex D. Don	Caprifoliaceae	Vibu cyli
178.	<i>Vitex negundo</i> L.	Verbenaceae	Vite negu
179.	<i>Zanthoxylum armatum</i> DC.	Rutaceae	Zant arma
180.	<i>Ziziphus mauritiana</i> Lam.	Rhamnaceae	Zizi maur

Shrubs were dominant with 63 species and followed by herbs (57 species), trees (35 species) and climbers (25 species). Angiosperms were dominant with 167 species (147 dicots and 20 monocots), gymnosperms were represented by 2 species and pteridophytes by 11 species (Table 3).

Table 3: Number of species in different plant groups and life-forms

S.N.	Plant group	Life-forms				Number of species
		Trees	Shrubs	Herbs	Climbers	
1.	Angiosperms	33	63	46	25	167
2.	Gymnosperms	2	-	-	-	2
3.	Pteridophytes	-	-	11	-	11
	Total	35	63	57	25	180

Out of the 68 families, Asteraceae reported the highest number of species (14 species), followed by Rosaceae (13 species), Fabaceae (10 species) and Liliaceae (9 species) (Table 4).

Table 4: List of families representing the 180 species

S.N.	Families	No. of species	No. of family
1.	Asteraceae	14	1
2.	Rosaceae	13	1
3.	Fabaceae	10	1
4.	Liliaceae	9	1
5.	Fagaceae, Pteridaceae	8	2
6.	Lauraceae	7	1
7.	Urticaceae	6	1
8.	Rubiaceae, Poaceae, Theaceae	5	3
9.	Myrsinaceae, Oleaceae	4	2
10.	Anacardiaceae, Berberidaceae, Cyperaceae, Ericaceae, Euphorbiaceae, Melastomaceae, , Ranunculaceae, Rutaceae	3	8

11.	Dryopteridaceae, Lamiaceae, Lardizabalaceae, Moraceae, Pinaceae, Polygonaceae, Rhamnaceae, Scrophulariaceae, Verbenaceae, Vitaceae, Zingiberaceae	2	11
12.	Acanthaceae, Actinidiaceae, Amaranthaceae, Apiaceae, Apocynaceae, Aquifoliaceae, Araliaceae, Aristolochiaceae, Asclepiadaceae, Athyriaceae, Betulaceae, Buxaceae, Cannabaceae, Caprifoliaceae, Celastraceae, Clusiaceae, Convallariaceae, Coriariaceae, Cornaceae, Dioscoreaceae, Flacourtiaceae, Gentianaceae, Geraniaceae, Gleicheniaceae, Hydrangeaceae, Hypoxidaceae, Juglandaceae, Menispermaceae, Myricaceae, Myrtaceae, Piperaceae, Primulaceae, Santalaceae, Sapotaceae, Solanaceae, Thymelaeaceae	1	36

4.3 Diversity indexes

Margalef index and Shannon-Wiener diversity index were found to be lowest (2.34, 1.90) and highest (3.18, 7.95) in plot 35 and plot 19 respectively. Pielou evenness was found to lowest (0.70) in plot 1 and highest (0.93) in plot 8 and plot 31. Simpson diversity index was lowest in plot 35 (0.80) and highest in plot 22 (0.95). The value of Margalef index, Shannon-Wiener diversity index, Pielou evenness index and Simpson diversity index in different plots sampled is shown in Appendix II.

Species richness was significantly correlated to Margalef index ($r = 0.97$, $p < 0.001$), Shannon-Wiener diversity index ($r = 0.81$, $p < 0.001$) and Simpson diversity index ($r = 0.46$, $p < 0.01$). However, the relationship with evenness was insignificant (Figure 3).

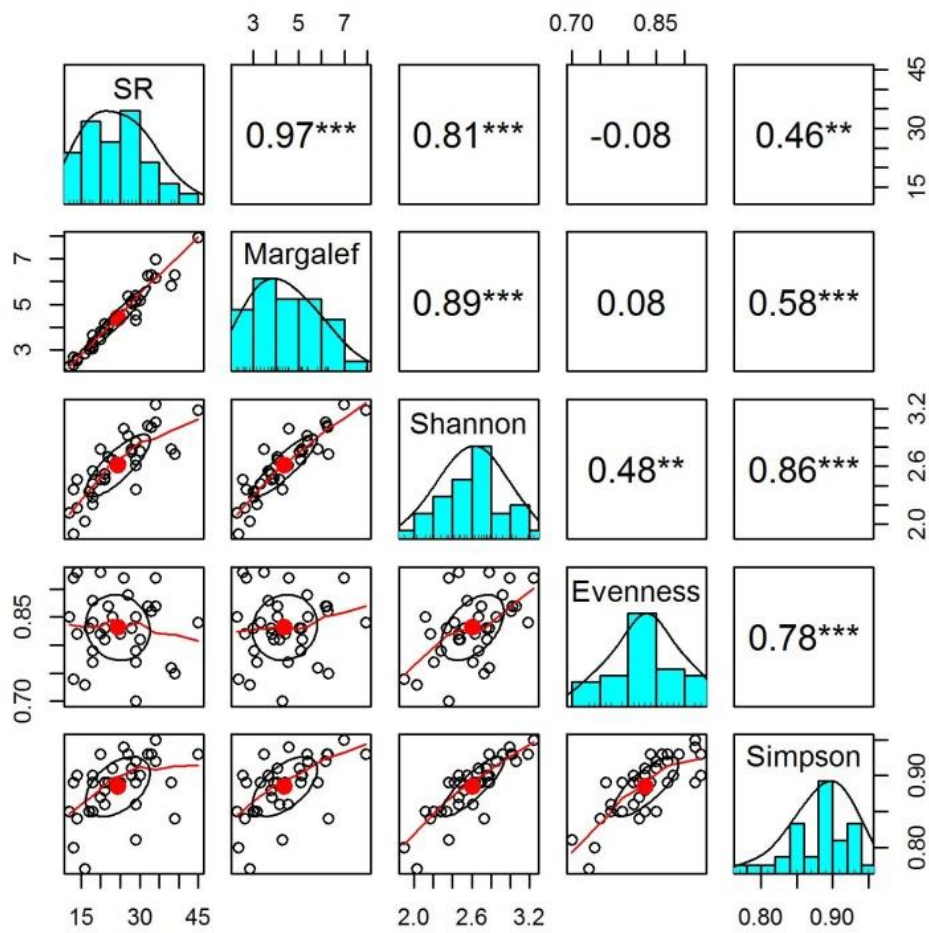


Figure 3: Correlation between species richness and diversity indexes (***) and ** indicate significant correlation at $p < 0.001$ and $p < 0.01$ respectively)

4.4 Soil chemical properties

Soil pH was found to be highest in plot 22 (6.25) and lowest in plot 27 (4.40). The highest and lowest value of TN was recorded in plot 12 (0.7 g/kg) and plot 9 (4.8 g/kg) respectively. For SOM, the highest and lowest value was recorded in plot 27 (126.40 g/kg) and plot 5 respectively (18.33 g/kg). The highest value (10.13 mg/kg) of AP was obtained in plot 30 and lowest (1.63 mg/kg) in plot 2, plot 12 and plot 18. The highest (208 mg/kg) value of AK was reported from plot 13, plot 29 and plot 30 while the lowest (19 mg/kg) value recorded from plot 28 (Table 5).

Table 5: Values of soil chemical properties in the plots sampled

Plot	pH	TN (g/kg)	SOM (g/kg)	AP (mg/kg)	AK (mg/kg)
1.	5.42	2.0	37.65	2.37	64
2.	5.62	2.3	56.48	1.63	97
3.	4.97	1.1	40.34	6.07	98
4.	4.99	1.1	28.24	3.11	42
5.	4.74	1.5	18.33	3.85	41
6.	4.96	1.5	52.44	5.33	41
7.	4.87	0.8	46.93	3.85	38
8.	4.92	2.2	65.89	2.37	59
9.	5.91	0.7	39.00	3.11	67
10.	4.79	2.7	69.93	3.85	200
11.	5.21	4.2	111.61	8.28	111
12.	4.87	4.8	125.06	1.63	202
13.	5.08	3.6	112.96	4.59	208
14.	4.75	1.5	63.20	2.74	98
15.	4.28	2.0	102.20	3.11	75
16.	4.58	2.8	104.89	4.59	106
17.	4.63	3.6	110.27	3.48	76
18.	5.14	3.6	98.16	1.63	155
19.	4.86	4.0	80.68	2.37	94
20.	4.90	3.4	118.34	4.59	104
21.	5.92	2.2	110.27	3.11	113
22.	6.25	3.1	91.44	5.33	104

23.	5.67	3.1	92.79	7.54	115
24.	4.29	4.0	104.22	8.65	122
25.	5.24	3.0	100.18	9.76	153
26.	5.91	4.5	105.56	4.96	110
27.	4.40	5.1	126.40	3.85	134
28.	4.99	4.6	55.13	6.07	19
29.	5.50	6.4	49.08	6.43	208
30.	4.78	3.4	116.32	10.13	208
31.	4.72	2.4	86.06	9.02	186
32.	5.15	1.9	67.24	6.07	123
33.	5.10	3.6	116.99	6.80	97
34.	5.07	2.4	80.68	6.80	132
35.	4.88	1.5	43.03	3.11	51

4.5 Correlation among different variables

Soil total nitrogen showed significant positive correlation with altitude ($r = 0.36$, $p < 0.05$). Species richness showed significant positive relation with pH ($r = 0.47$, $p < 0.01$). Among the various life-forms studied, significant correlation of shrub ($r = 0.42$, $p < 0.05$), herb ($r = 0.54$, $p < 0.001$) and climber ($r = 0.41$, $p < 0.05$) was found with soil pH. However, the tree was not significantly related to soil pH. The relationship of species richness with aspect, slope, TN, SOC, AP and AK were found to be not significant (Figure 4). Among the soil variables, TN and SOM were found to be significantly and positively correlated ($r = 0.58$, $p < 0.001$). AK showed significant correlation with both TN ($r = 0.51$, $p < 0.01$) and SOM ($r = 0.52$, $p < 0.01$)

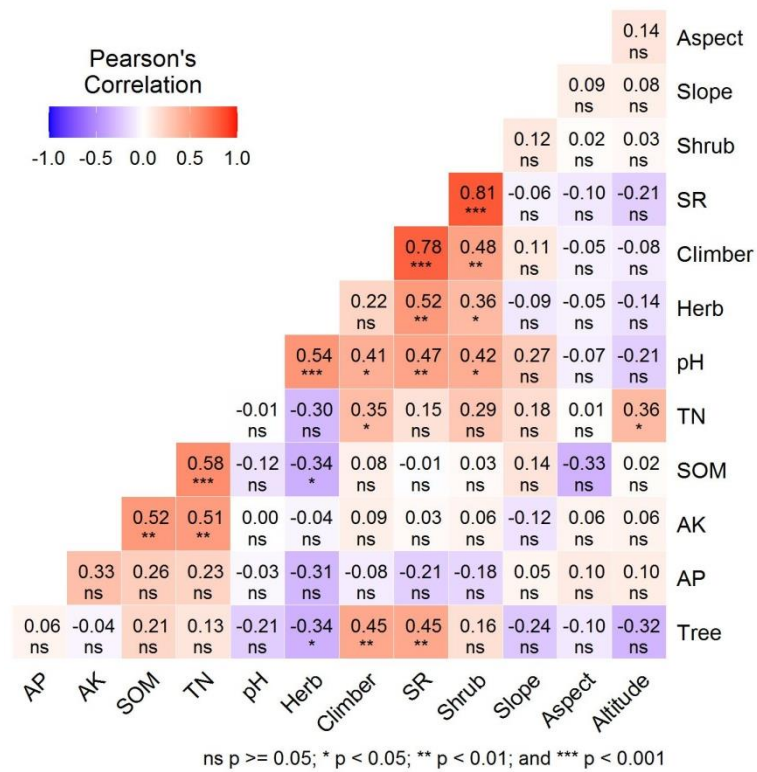


Figure 4: Correlation between species richness and environmental variables

4.6 ANOVA result

Only species richness was found significantly different along soil pH. Species richness was not found significantly different along altitude, aspect, slope, TN, SOM, AP and AK (Table 6).

Table 6: One-way ANOVA result

Environmental factors	Variables	F value	p value
Topographic factors	Altitude	1.301	0.262
	Aspect	0.269	0.607
	Slope	0.091	0.765
Soil factors	pH	8.330	0.007**
	TN	0.874	0.357
	SOM	0.016	0.901
	AP	1.500	0.229
	AK	0.007	0.935

** indicate significant difference at $p < 0.01$

4.7 Relationship of species richness with topography and soil

The model was chosen using Akaike Information Criterion (AIC) in a stepwise algorithm. Four variables were found to be significantly associated with species richness (Table 7).

Table 7: Summary of model selection

Variables	AIC	Coefficients	Standard error	z value	Pr (> z)
Slope	246.80	-0.007	0.003	-1.999	0.04562*
AP	247.91	-0.035	0.016	-2.238	0.02523*
TN	250.71	0.082	0.029	2.805	0.00503**
pH	261.15	0.324	0.075	4.310	0.00002***

***, ** and * indicate significant correlation at $p < 0.001$, $p < 0.01$ and $p < 0.05$ respectively

Each significant environmental variable (slope, AP, pH, TN) obtained after model selection was then tested for first order linear model and second order polynomial. Soil pH was found to be the best variable explaining species richness (Table 8).

Table 8: Regression statistics for species richness against environmental parameters

Variables	Model	Order	Residual d.f	Residual deviance	d.f	Deviance	F	Pr (>F)
Slope	Null	0	34	93.034				
	GLM	1	33	92.748	1	0.286	0.101	0.753
	GLM	2	32	92.693	2	0.341	0.058	0.944
pH	Null	0	34	93.034				
	GLM	1	33	72.791	1	20.242	8.834	
	GLM	2	32	72.749	2	20.285	4.284	0.006** 0.023*
TN	Null	0	34	93.034				
	GLM	1	33	90.857	1	2.177	0.775	0.385
	GLM	2	32	90.773	2	2.261	0.390	0.680
AP	Null	0	34	93.034				
	GLM	1	33	88.997	1	4.037	1.479	0.233
	GLM	2	32	84.084	2	8.950	1.695	0.199

** and * indicate significant correlation at $p < 0.01$ and $p < 0.05$ respectively

The result of (Generalized Linear Model) GLM revealed the linear pattern of species richness of different life-form along soil pH. Species richness ($R^2 = 0.227$, $p = 0.005$), shrubs ($R^2 = 0.177$, $p = 0.015$), herbs ($R^2 = 0.252$, $p = 0.001$) and climber ($R^2 = 0.159$, $p = 0.018$) showed positive trend with soil pH. However, no significant relation was observed between trees ($R^2 = 0.052$, $p = 0.223$) and soil pH (Table 9, Figure 5).

Table 9: Statistics obtained after testing of response variables along soil pH with different order model using GLM

Response	Model	Order	Residual d.f	Residual deviance	d.f	Deviance	F	Pr (>F)
SR	Null	0	34	93.034				
	GLM	1	33	72.791	1	20.242	8.834	0.005**
	GLM	2	32	72.749	2	20.285	4.284	0.022*
Tree	Null	0	34	53.466				
	GLM	1	33	51.107	1	2.360	1.581	0.217
	GLM	2	32	48.335	2	5.131	1.697	0.199
Shrub	Null	0	34	58.122				
	GLM	1	33	48.292	1	6.352	6.352	0.017*
	GLM	2	32	48.215	2	3.116	3.116	0.058
Herb	Null	0	34	71.905				
	GLM	1	33	53.127	1	12.454	12.45	0.001**
	GLM	2	32	49.120	2	22.785	4	0.002**
Climber	Null	0	34	51.901				
	GLM	1	33	44.272	1	7.630	5.970	0.020*
	GLM	2	32	43.274	2	8.627	3.300	0.049*

** and * indicate significant correlation at $p < 0.01$ and $p < 0.05$ respectively

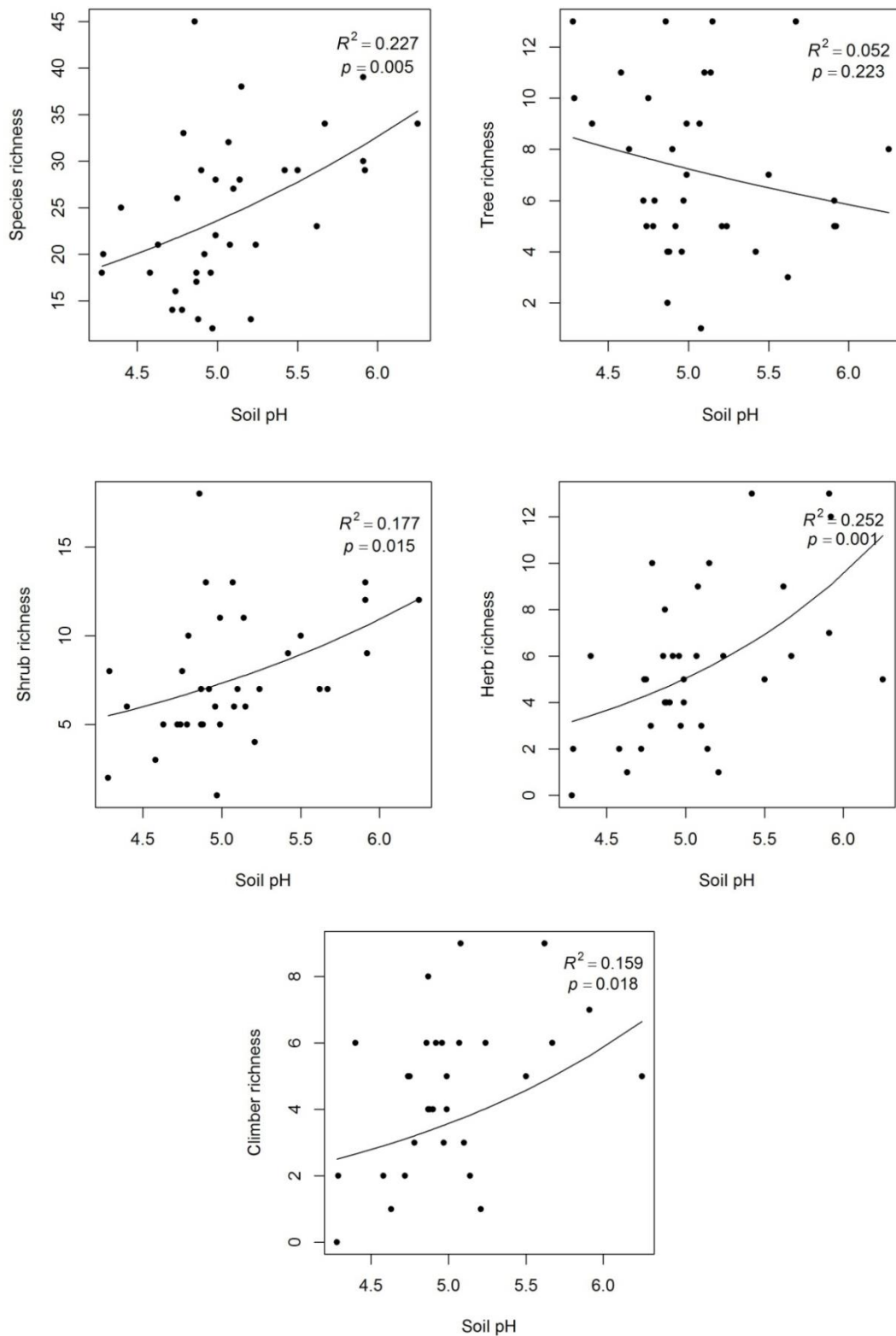


Figure 5: Plots showing species richness relationship with soil pH (line fitted with GLM first order)

Species richness showed positive correlation with TN and AK while negative correlation with SOM and AP. However, none of the nutrients showed significant

relation. Among the different life-forms studied, only herb and climber showed significant negative and positive correlation with SOM ($R^2 = 0.114$, $p = 0.046$) and TN ($R^2 = 0.122$, $p = 0.045$) respectively (Figure 6).

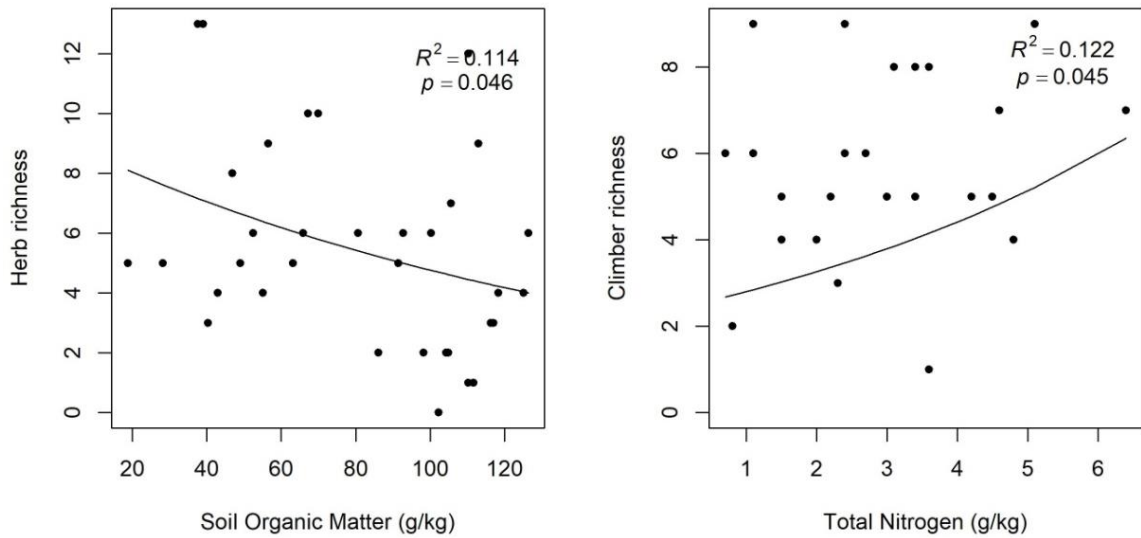


Figure 6: Plots showing herb richness and climber richness relationship with soil SOM and TN (line fitted with GLM first order)

4.8 Mean decrease accuracy and importance value

The mean decrease accuracy tells about the decrease in model accuracy if a variable is left out. In our model, pH had the highest mean decrease accuracy which means if it was left out, the accuracy of the model would decrease. Variable importance was used to rank the importance of different environmental variables. Importance value of pH was found to be highest and followed by aspect, altitude, AP, TN, SOM, AK and slope. Both the results of variable importance and mean decrease accuracy confirmed that soil pH was the strongest environmental variable in our study (Table 10, Figure 7).

Table 10: Variable importance and mean decrease accuracy of different environmental variables

Variables	Importance	%IncMSE
pH	100.000	7.046
Aspect	59.981	-0.238
Altitude	46.318	0.322
AP	36.213	2.205
TN	9.701	3.616
SOM	8.175	4.592
AK	2.575	2.947
Slope	0.000	3.097

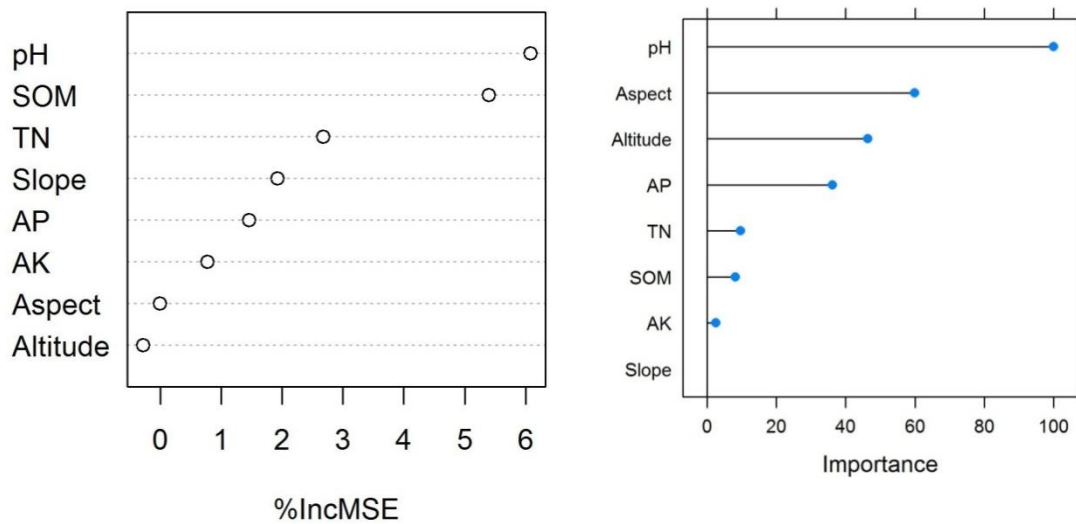


Figure 7: Plot showing mean decrease accuracy (%IncMSE) and importance of different environmental variables

4.9 Environment-species interaction

The result obtained from the Detrended Correspondence Analysis (DCA) of species data against the plots (Table 11) showed the axis length one is 2.5 greater than standard deviation. Thus, CCA was performed.

Table 11: DCA ordination summary

	DCA1	DCA2	DCA3	DCA4
Eigenvalues	0.479	0.450	0.249	0.169
Additive Eigenvalues	0.479	0.450	0.248	0.170
Decorana values	0.552	0.388	0.207	0.155
Axis lengths	5.129	2.997	2.574	1.779

CCA axis 1 was positively related with altitude, AP, AK and negatively correlated to aspect. CCA axis 2 was positively related to pH and negatively to TN, slope and SOM. CCA1, CCA2, CCA3 and CCA4 axes were found to be highly correlated to altitude, TN, pH and AK respectively (Table 12).

Table 12: Correlation coefficients of environmental variables with CCA axes

	CCA1	CCA2	CCA3	CCA4
Altitude	0.909	0.070	0.058	0.365
Aspect	-0.094	0.174	0.263	0.258
Slope	0.059	-0.313	-0.092	0.296
pH	-0.199	0.150	-0.851	-0.280
TN	0.515	-0.752	-0.110	0.038
SOM	0.330	-0.701	0.149	-0.303
AP	0.270	-0.099	0.378	-0.181
AK	0.392	-0.314	0.179	-0.761

From the CCA ordination, 33.43 % of the variation is explained by environmental variables. First CCA axis was found to explain 23.37 % of the variation, the second CCA axis explained 16.81 %, third CCA axis explained 16.16 % and fourth CCA axis explained 13.21 % of the variation. Hence, a total of 69.54 % of the variation found to be explained by these four axes (Table 13).

Table 13: Percentage of variation explained by CCA axes

Constrained inertia	CCA axes	Eigen values	Percentage variation explained	Cumulative variation %
2.0689	CCA1	0.4835	23.370	
	CCA2	0.3477	16.810	40.170
	CCA3	0.3343	16.160	56.330
	CCA4	0.2733	13.210	69.540
	CCA5	0.1861	8.993	78.538
	CCA6	0.1692	8.178	86.715
	CCA7	0.1487	7.189	93.964
	CCA8	0.1261	6.096	100.000

Altitude, pH, TN and AK were found to be the significant variables affecting species composition (Table 14). Along the CCA axis 1, the distribution of species were found to be affected by altitude where as along CCA axis 2 they were found to be influenced by TN. The closer the species, the similar are their requirements. The CCA plot showed how species were distributed along the environmental variables. The length of the arrow indicates the correlation between the environmental variables and the species (Figure 8).

Table 14: Effect of environmental variables on species composition

Variable	Df	Chi square	F	Pr (>F)
Altitude	1	0.444	2.803	0.001***
Aspect	1	0.206	1.302	0.127
Slope	1	0.202	1.274	0.092
pH	1	0.307	1.940	0.001***
TN	1	0.311	1.967	0.010**
SOM	1	0.198	1.250	0.142
AP	1	0.170	1.074	0.299
AK	1	0.228	1.442	0.040*
Residual	26	4.119		

***, ** and * indicate significant correlation at $p < 0.001$, $p < 0.01$ and $p < 0.05$ respectively

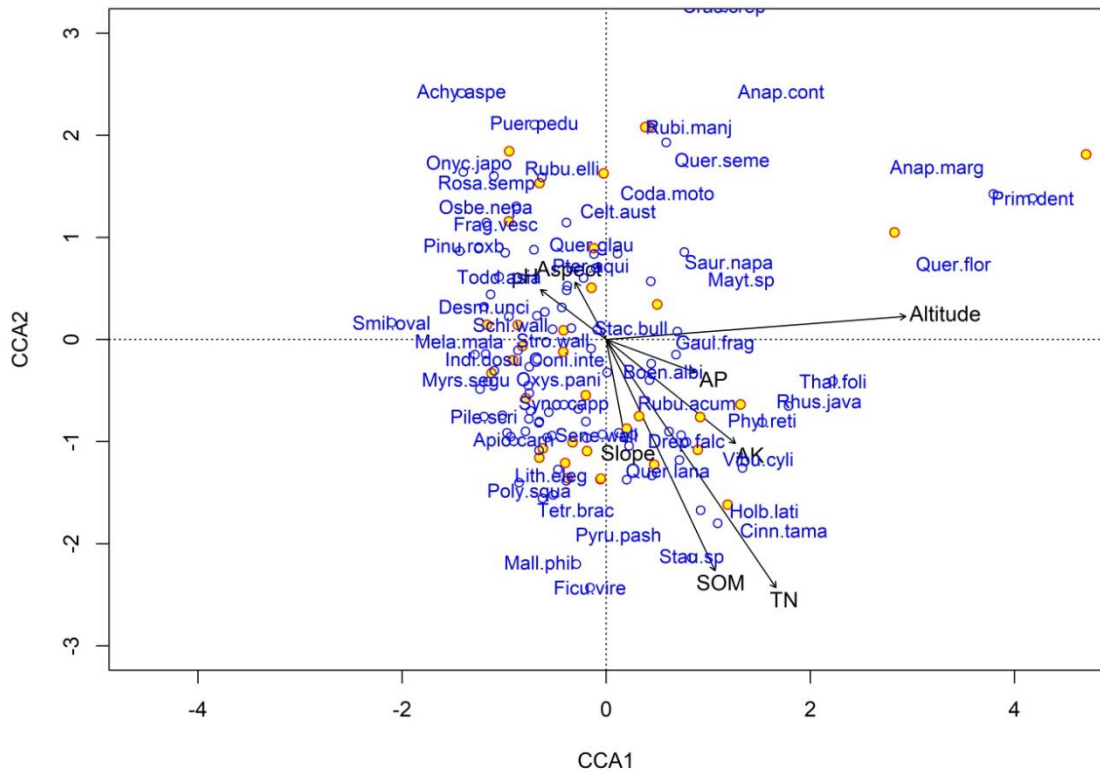


Figure 8: CCA plot showing the effect of environmental variables on species composition (the arrows represent the predictor variable, the circle represents the plot and the abbreviations are the names of the species)

Species like *Anaphalis margaritacea*, *Berberis aristata*, *Berberis wallichiana*, *Bidens pilosa*, *Capillipedium assimile*, *Neocinnamomum caudatum*, *Carex pendula*, *Cornus oblonga*, *Daphne bholua*, *Eriobotrya dubia*, *Gaultheria fragrantissima*, *Gentiana capitata*, *Gerbera maxima*, *Ilex aquifolium*, *Osbeckia nepalensis*, *Pinus roxburghii*, *Piptanthus nepalensis*, *Primula denticulata*, *Quercus floribunda*, *Randia tetrasperma*, *Rhus chinensis*, *Rhus javanica*, *Rubus acuminatus*, *Rubus ulmifolius*, *Saurauia napaulensis*, *Schima wallichii*, *Smilax aspera*, *Smilax elegans*, *Smilax ferox*, *Smilax lanceifolia*, *Smilax ovalifolia*, *Viburnum cylindricum* are highly correlated with altitude.

Soil pH strongly affected *Achyranthes aspera*, *Apios carnea*, *Castanopsis tribuloides*, *Chromolaena odorata*, *Clematis connata*, *Dicranopteris linearis*, *Dioscorea deltoidea*, *Elatostema lineolatum*, *Eurya cerasifolia*, *Hydrangea febrifuga*, *Jasminum officinale*, *Juglans regia*, *Maesa chisia*, *Melastoma malabathricum*, *Molinieria capitulata*, *Myrsine seguinii*, *Neolitsea sericea*, *Onychium japonicum*, *Osyris wightiana*, *Persea duthiei*, *Prunus cerasoides*, *Pteris vittata*, *Quercus glauca*, *Rhus*

wallichii, *Roscoea purpurea*, *Sarcococca hookeriana*, *Smilax glauca*, *Strobilanthes wallichii*.

Soil TN showed major effects on *Ageratina adenophora*, *Ainsliaea latifolia*, *Albizia* sp., *Anaphalis contorta*, *Artemisia indica*, *Camellia kissi*, *Celtis australis*, *Cheilanthes farinosa*, *Codariocalyx motorius*, *Crassocephalum crepidioides*, *Drepanostachyum falcatum*, *Eragrostis tenella*, *Galium mollugo*, *Homalium napaulense*, *Hoya lanceolata*, *Lithocarpus elegans*, *Lyonia ovalifolia*, *Machilus odoratissima*, *Mallotus philippensis*, *Myrsine capitellata*, *Myrsine semiserrata*, *Ophiopogon* sp., *Pinus wallichiana*, *Polystichum squarrosus*, *Pteridium aquilinum*, *Pueraria peduncularis*, *Pyrus pashia*, *Quercus semecarpifolia*, *Quercus lanata*, *Rubia manjith*, *Rubus ellipticus*, *Rubus paniculatus*, *Smilax perfoliata*.

CHAPTER FIVE

5. DISCUSSION

5.1. Species diversity

Species richness is a reliable parameter for assessing biodiversity. Sites with more species are considered richer, hence are important from ecological and biodiversity point of view. Dani and Baniya (2022) in a previous study in the same site reported 47 tree species which is higher than the 35 tree species recorded in the present study. Previous studies (Bhattarai and Vetaas 2003, Chawla et al. 2008) on species richness reported higher number of species in comparison to the present study. This might be due to the variation in the number of plots sampled and altitudinal gradient studied.

5.2 Diversity parameters

All plots except one showed value below two otherwise in Shannon-Weiner diversity index was above two. Low value indicates low diversity and high value high diversity. Higher the Pielou evenness value, higher is the uniformity in the number of individuals of species. In present study most of the values were close to one which indicated the uniform distribution of individuals within species. Simpson diversity index is less sensitive to rare species and gives more importance to common species. Higher its value, higher is the diversity. Simpson diversity values were near to one indicating high diversity in our study site.

5.3 Correlation among different variables

Altitude showed significant positive correlation with TN. With increase in altitude, the microbial activities get retarded, as a result of which there is reduction in litter decomposition and most of the nutrients remain bounded in soil and not utilized by plants. This ultimately leads to higher accumulation of nitrogen in soil.

In the present study, species richness was positively correlated to TN and AK but negatively correlated to SOM and AP. Nitrogen is a limiting factor in temperate forests so that might be the reason for positive relations as increase in nitrogen concentration enhances the growth of those species that otherwise are stressed due to

its limitation. Potassium does not combine with organic compounds and is easily leached from the soil. Hence, its availability due to lesser leaching might have resulted in the positive correlation. SOM is the major source of nutrients for plants. The nutrients released by it through the process of decomposition are the main source of nutrition. But slow decomposition might be the reason for its negative effect on species richness as deficiency of nutrients hampers plant growth and development. Phosphorus is reported to be a limiting factor in tropical forests but our study is in temperate forests so it might not be limiting. The increase in phosphorus might favor fewer competitive species which might have resulted in the negative relation.

TN showed significant positive correlation with AK which is in agreement with other studies (Gupta and Sharma 2008). The availability of nitrogen is dependent on SOM. AK also showed a significant positive correlation with SOM. A layer of organic matter is found to improve retention of K in soils (Boruah and Nath 1992).

5.4. Influence of topography on species richness

Among the topographic factors, most of the studies had reported altitude as the most influential one influencing species richness (Teshome et al. 2020). In the present study species richness showed negative correlation with altitude. Previous studies also showed decrease in species richness with increase in altitude (Saikia et al. 2017, Sharma et al. 2009, Teshome et al. 2020). In contrast many studies reported a peak in species richness at mid altitude (Bhattarai and Vetaas 2003, Chawla et al. 2008, Das et al. 2020, Sharma et al. 2019, Song and Cao 2017). With increase in altitude diversity decreases and few species remain at extreme altitude (Pavón et al. 2000). The decrease in species richness with altitude could be due to eco-physiological constraints, low temperature, short growing season and productivity (Körner 1998). At high altitude, plant growth as well as microbial activity is affected by low temperature. Also, the biogeochemical cycle is retarded by low temperature which limits availability of nutrients. Low temperature also reduces water conductivity which affects nutrient consumption as movement of nutrients to roots from soil take place through water flow (Cornwell and Grubb 2003). The overlapping of habitat at mid-points in comparison to the periphery might promote the co-existence of a large number of species at mid-point resulting in hump-shaped patterns.

Most of the previous studies depicted inverse correlation between species richness and degree of slope (Sharma et al. 2009, Teshome et al. 2020, Yang et al. 2021) which could be due to the steepness. No significant trend was observed between species richness and aspect. Slope affects species richness by influencing soil properties. High degree of slope results in greater surface run-off accompanied by translocation of surface materials down slope through soil erosion and movement of the soil mass (Heydari and Mahdavi 2009). This surface run-off and erosion creates deficiency of nutrients and substrate for plant growth.

Aspect influences species richness by controlling the incoming radiation. No significant relation was observed in the present study between species richness and aspect which was similar to the by Teshome et al. (2020), Yang et al. (2021). In present study, north-west facing slope showed highest species richness while south-east facing slope reported lowest species richness. Most of the studies (Yang et al. 2020) done in the past reported higher species richness in the north facing slope as compared to south facing slope. South-facing slopes are much drier which favors the growth of drought-tolerant species. Moreover, less moisture in soil might limit the absorption of nutrients by plants as plant nutrient uptake occurs through soil solution. In north-facing slopes vegetation is dense and thick whereas south-facing slopes have scattered and thin vegetation (Singh 2018).

Topographic factors showed no significant influence on species richness in our study. These factors might have indirectly affected the species richness by influencing the soil properties. Topography influences species richness by controlling incoming solar condition, soil moisture, microbial activity, temperature, litter decomposition. At higher elevation low temperature causes slow decay of organic matter resulting in accumulation of humus but limits soil productivity and nutrient absorption. Altitude influences species richness by creating variation in climate but in present study the difference in climate along the elevation gradient might not have varied markedly to significantly influence species. Moreover, present study was at local level and at regional level climate is reported to be the deciding factor. At the local level, environmental heterogeneity overpowers in influencing species. The coexistence of a greater number of species is affected by local variations in the relative amount of nutrients, soil physical characteristics and biotic factors, when temperature and moisture are not acting as limiting factors (Sánchez-González and Looper-Mata 2005).

5.5 Influence of soil properties on species richness

In our study, soil pH was found to be the important factor affecting species richness which is supported by the result of Dinggaan et al. 2017. Species richness showed significant positive relation with pH which is in agreement with other studies (Bhattarai et al. 2014, Riesch et al. 2018, Roem and Berendse 2000, Stevens et al. 2004, Vetaas 1997, Weiher et al. 2004). Shrub, herb and climber also showed significant positive trends with soil pH as total species richness. No significant trend was observed between soil pH and tree. Some studies also reported negative correlation between species richness and pH (Diamond and Smeins 1995, Palpurina et al. 2017) while others a unimodal response. At very low pH, nutrients become less available to plants and at a certain pH level most desirable nutrients absorption takes place in plants (McFarland et al. 2015). The values of mean decrease accuracy and variable importance also confirmed the soil pH as an important variable influencing species richness in our study.

Soil pH plays an important role in controlling soil nutrients availability. At slightly acidic pH, the maximum availability of all soil nutrients at plant roots is reported (Taiz & Zeiger 2002). Most species are incapable of acquiring nutrients outside their optimum pH range i.e. higher or lower than optimum range (Schuster and Diekmann 2003). The imbalance of nutrients affects growth and development of plants (Khadka et al. 2016). The availability of nutrients is dependent on soil pH as nutrients are released after decomposition by microbes and populations of microbes are affected by soil pH. Soil nutrients affect the species. In low-nutrient soils, high plant diversity is possible due to ability of soil to limit the growth of the highly competent species that can outcompete and exclude other species, hence promoting the coexistence of numerous species (Pena-Claros et al. 2012).

Forest soil needs to be slightly acidic for nutrient supply to be balanced. Through soil microbial activity, soil pH indirectly controls nitrogen availability (Ahmad et al. 2011). Low pH retards microbial activity as well as slows down the processes of nitrogen mineralization and nitrification (Kimura et al. 2009). Similarly soil pH influences availability of phosphorus as for soil having pH between 5.3 and 5.5 maximum amount of phosphorus was recorded. In case of potassium, maximum amount was recorded between pH 5.0 to 5.5 (Malik and Haq 2022).

Our findings reported no significant effect of soil nutrients on total species richness. However, herbs and climbers succeeded in showing some significant relation with SOM and TN respectively. Herb richness decreased with increase in SOM. SOM affects species by having an important influence on soil physical and chemical properties, soil fertility status, plant nutrition and biological activity in the soil (Syaed 2021). Accumulation of SOM and not its conversion to usable forms required by plants might be the reason for decrease in herb richness. Climber richness increased with increase in TN. Climber needs support for their growth and trees can play that role very efficiently. Trees being woody and giant require high amount of nutrients for their growth. The higher the nutrients the more will be the growth of the trees and support for the climbers. This could be the reason for increase in climber richness with increase in TN.

Previous studies suggested a hump-shaped pattern between soil nutrients and species richness (Audet et al. 2015, Pausas and Austin 2001). At intermediate levels, species richness is found to be highest. Higher nutrient levels increase the dominance of few competitive species as a result decline in species richness occurs. Similarly at low nutrients, the plant growth is disturbed due to nutrient limitation. Thus, at intermediate nutrient levels, species richness becomes high as no species becomes dominant as well as nutrients are available for plant growth and development.

At local level the species richness is determined by the resource availability and environmental variables that have direct effect on plant growth and development (Pausas and Austin 2001). Due to the interactive and dynamic nature of soil variables, it is difficult to distinguish the local effects of soil physical and chemical properties on species richness. Organic matter determines the stability of soil aggregates, porosity, gas and water exchange and availability of nutrients (Schoenholtz et al. 2000).

5.6. Species composition

CCA was used to evaluate the relationship between species richness and environmental variables. The closer the species in CCA plot, the more resemblance in their niche and needs. The smaller the distance among the species, the higher is their correlation. From the results of CCA, it was found that altitude and pH were the factors strongly affecting species composition in the study area. The composition of species is chiefly determined by elevation (Rai et al. 2016, Villanueva and Bout 2018)

and the related environmental factors (Culmsee and Leuschner 2013). With increase in altitude, temperature decreases. Low temperature also affects the microbial activity of soil microbes, decomposition process and productivity. With the increase in elevation, the environmental conditions become stressful as productivity is limited and nutrient cycling becomes slow. Only plants that can adapt to cold and stressful conditions are found at higher elevations. So, altitude is a major deciding factor governing the distribution of species through its influence on growing season, temperature, moisture, productivity (Shaheen et al. 2015).

Soil pH affects species composition (Chandra et al. 2018) by affecting the availability of nutrients. Plants are not adapted to absorb nutrients at all pH. Plants vary in their tolerance to acidity and basicity as well as nutrients requirements. Most plants have an optimum range within which most of the nutrients required by plants are absorbed. At extremely acidic and alkaline soils, plant nutrients become unavailable to plants due to different reactions in soil which transform them into a form unavailable to plants. Hence soil pH affects species composition as plants varies in their tolerance to acidity and basicity and as well as requirements of nutrients.

Among the various soil nutrients analyzed, TN and AK were found to significantly influence species composition. Soil nutrients are essential for plant growth and reproduction and greatly influence species composition (Becknell and Powers 2014, Idowu et al. 2020, Long et al. 2018, Yang et al. 2021). Nitrogen is an essential element for plant growth and development. In temperate forest, nitrogen is found to be a limiting factor (Tilman et al. 1996). Its deficiency causes imbalance in plant growth and development. Soil total nitrogen is majorly present in soil organic matter (Quan et al. 2014). As a result, its availability is influenced by accumulation and decomposition of organic matter (Sokolov et al. 2008).

The effect of edaphic factor is found to be stronger than that of topographic factors in influencing species composition. The distribution of species is determined by the interaction of multiple environmental variables.

CHAPTER SIX

6. CONCLUSION

The present study was focused in order to identify the effect of environmental variables on species diversity. Altogether 180 species were reported from the 35 plots sampled. Species richness showed decreasing pattern with both elevation and slope. Species richness found to be increasing with increasing soil pH. On the other hand, species richness showed positive relation with some nutrients and negative relation with others. So our hypothesis of hump-shaped relation between species richness and different environmental got rejected. In the present study, soil pH found to be the most influential factor affecting species richness. Species richness, shrub, herb and climber reported to be increasing with soil pH. At local level, soil plays a major role in governing species richness. Species composition was significantly affected by altitude, TN, pH and AK. Hence, in the present study species richness was prominently affected by soil pH whereas the interaction of multiple factors affected species composition in the study area.

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Appendix I: Physical characteristics of the plots in study area

S. N.	Location	Altitude (m)	Slope	Aspect	Latitude	Longitude
1.	Godam	1636	12°	82°	27° 41.117 N	85° 12.250 E
2.	Godam	1740	37°	169°	27° 41.183 N	85° 12.183 E
3.	Godam	1829	33°	151°	27° 41.250 N	85° 12.167 E
4.	Godam	1930	22°	68°	27° 41.314 N	85° 12.033 E
5.	Godam	2036	24°	151°	27° 41.289 N	85° 11.889 E
6.	Godam	2133	25°	73°	27° 41.267 N	85° 11.733 E
7.	Godam	2230	35°	107°	27° 41.064 N	85° 11.562 E
8.	Godam	2330	10°	43°	27° 41.022 N	85° 11.409 E
9.	Machhegaun	1527	12°	4°	27° 39.392 N	85° 15.033 E
10.	Machhegaun	1637	28°	47°	27° 39.333 N	85° 14.774 E
11.	Machhegaun	1727	35°	69°	27° 39.362 N	85° 14.637 E
12.	Machhegaun	1828	41°	74°	27° 39.334 N	85° 14.568 E
13.	Machhegaun	2417	17°	11°	27° 39.036 N	85° 14.085 E
14.	Laglage	1550	24°	155°	27° 41.797 N	85° 12.212 E
15.	Laglage	1650	15°	57°	27° 41.871 N	85° 12.049 E
16.	Laglage	1751	10°	20°	27° 41.848 N	85° 11.863 E
17.	Laglage	1851	27°	42°	27° 41.726 N	85° 11.782 E
18.	Laglage	1951	27°	11°	27° 41.642 N	85° 11.663 E
19.	Laglage	2051	27°	300°	27° 41.509 N	85° 11.607 E
20.	Laglage	2149	34°	41°	27° 41.310 N	85° 11.613 E
21.	Chandragiri	1543	35°	14°	27° 41.049 N	85° 12.872 E
22.	Chandragiri	1645	39°	25°	27° 41.003 N	85° 12.817 E

23.	Chandragiri	1741	30°	48°	27° 41.946 N	85° 12.787 E
24.	Chandragiri	1839	22°	49°	27° 41.881 N	85° 12.722 E
25.	Chandragiri	1940	37°	359°	27° 41.681 N	85° 12.667 E
26.	Chandragiri	2039	42°	10°	27° 41.616 N	85° 12.670 E
27.	Chandragiri	2141	20°	48°	27° 41.359 N	85° 12.500 E
28.	Chandragiri	2241	50°	14°	27° 41.260 N	85° 12.446 E
29.	Chandragiri	2338	12°	313°	27° 41.159 N	85° 12.398 E
30.	Chandragiri	2440	29°	86°	27° 41.034 N	85° 12.384 E
31.	Matatirtha	1529	13°	110°	27° 41.680 N	85° 13.755 E
32.	Matatirtha	1628	20°	78°	27° 41.662 N	85° 13.599 E
33.	Matatirtha	1729	27°	38°	27° 41.642 N	85° 13.436 E
34.	Matatirtha	1830	15°	93°	27° 41.438 N	85° 13.271 E
35.	Matatirtha	1929	38°	345°	27° 41.636 N	85° 13.066 E

Appendix II: Species richness and diversity parameters of the plots sampled

Plot	Tree	Shrub	Climber	Herb	Total	M	H	E	D
1.	4	9	3	13	29	4.29	2.36	0.70	0.81
2.	3	7	4	9	23	4.07	2.67	0.85	0.91
3.	6	1	2	3	12	2.28	2.12	0.85	0.85
4.	9	5	4	5	23	4.07	2.70	0.87	0.89
5.	5	5	1	5	16	2.85	2.03	0.73	0.77
6.	4	6	2	6	18	3.05	2.28	0.79	0.85
7.	2	5	2	8	17	3.02	2.34	0.83	0.85
8.	5	7	2	6	20	3.46	2.78	0.93	0.93
9.	6	13	4	13	36	6.29	2.73	0.75	0.84
10.	6	10	5	10	31	6.28	3.01	0.86	0.93
11.	5	4	3	1	13	2.69	2.36	0.92	0.89
12.	4	7	3	4	18	3.66	2.55	0.88	0.90
13.	1	6	4	9	20	4.15	2.46	0.81	0.86
14.	10	8	4	5	27	4.55	2.99	0.92	0.94
15.	13	2	2	-	17	3.32	2.42	0.84	0.89
16.	11	3	2	2	18	3.15	2.21	0.77	0.85
17.	8	5	7	1	21	3.91	2.48	0.81	0.89
18.	11	11	4	2	28	5.13	2.78	0.83	0.90
19.	13	18	8	6	45	7.95	3.18	0.84	0.93
20.	8	13	4	4	29	5.10	2.67	0.79	0.89
21.	5	9	3	12	29	4.54	2.61	0.77	0.86
22.	8	12	7	5	32	6.98	3.24	0.92	0.95

23.	13	7	8	6	34	6.18	3.06	0.87	0.92
24.	10	8	-	2	20	3.81	2.46	0.82	0.87
25.	5	7	3	6	21	3.78	2.52	0.83	0.88
26.	5	12	5	7	29	5.16	2.76	0.81	0.90
27.	9	6	4	6	25	4.42	2.63	0.82	0.89
28.	7	11	6	4	28	5.04	2.75	0.83	0.91
29.	7	10	7	5	29	5.40	2.85	0.85	0.92
30.	5	5	1	3	14	2.62	2.17	0.82	0.84
31.	6	5	1	2	14	2.54	2.46	0.93	0.90
32.	13	6	9	10	38	5.83	2.78	0.76	0.89
33.	11	7	5	3	26	5.36	2.92	0.89	0.93
34.	9	13	4	6	32	6.25	3.02	0.87	0.93
35.	4	5	-	4	13	2.34	1.90	0.74	0.80

Note: **M** - Margalef index, **H** - Shannon-Weiner diversity index, **E** – Pileou’s evenness and **D** - Simpson diversity index

Appendix III: CCA summary

a. Species scores

Species	CCA1	CCA2	CCA3
Achy aspe	-0.986	1.420	-1.459
Adia capi	-0.833	0.189	-0.858
Ager aden	-0.271	0.675	-0.138
Ains lati	0.547	-0.591	-0.044
Albi sp	0.150	-2.170	0.312
Alnu nepa	-0.530	-0.266	0.596
Anap cont	1.190	1.423	0.305
Anap marg	2.253	0.986	-0.107
Apio carn	-0.630	-0.588	-1.964
Aris serp	-0.132	-0.566	0.075
Arte indi	-0.438	0.936	-0.874
Arun nepa	-0.974	0.971	0.939
Aspa seta	-0.999	0.509	-0.905
Athy fili	-0.999	0.509	-0.905
Berb aris	0.531	0.505	0.002
Berb wall	2.637	0.843	1.443
Bide pilo	2.905	0.8166	-0.234
Boeh nive	-0.999	0.509	-0.905
Boeh plat	-0.524	-0.157	-0.011
Boen albi	0.388	-0.184	-0.157
Budd asia	-0.391	-0.419	1.281
Came kiss	-0.361	-0.896	0.235
Capi assi	2.905	0.816	-0.234
Care bacc	-0.265	0.311	0.349

Care pend	2.905	0.816	-0.234
Care sp	-0.630	-0.588	-1.964
Cast indi	-0.072	0.413	0.558
Cast trib	-0.456	-0.472	0.586
Celt aust	0.080	0.741	-0.314
Chei fari	0.802	1.918	0.508
Cinn tama	1.210	-1.100	-0.209
Clem vita	0.582	-1.261	-0.506
Clem conn	-0.630	-0.588	-1.964
Coda moto	0.417	0.847	-0.407
Coni inte	-0.268	-0.114	-0.053
Cori nepa	0.075	0.497	0.535
Corn oblo	-0.515	-0.411	0.249
Cras crep	0.802	1.919	0.508
Crot cyti	0.075	0.497	0.535
Daph bhol	0.928	-0.742	-0.835
Desm unci	-0.833	0.189	-0.858
Desm sp	-0.110	-1.430	1.834
Dicr line	-0.492	0.519	1.340
Dios delt	-0.986	1.420	-1.459
Dode gran	-0.310	-0.419	1.281
Drep falc	0.546	-0.595	-0.071
Elat line	-0.458	-0.482	-1.297
Erag tene	0.407	1.138	0.177
Erio dubi	-0.366	0.059	-0.115
Eury acum	0.180	-0.550	0.448
Eury cera	-0.601	-0.062	0.877
Ficu otto	-0.676	-0.539	-0.720

Ficu vire	-0.110	-1.430	1.834
Frag vesc	-0.753	0.655	-1.048
Gali moll	0.310	1.218	-1.051
Gaul frag	0.741	-0.026	0.220
Gent capi	2.905	0.816	-0.234
Gera nepa	-0.999	0.509	-0.905
Gerb maxi	2.905	0.816	-0.234
Gira dive	-0.999	0.509	-0.905
Hede nepa	0.484	0.045	-0.841
Hedy sp	-0.833	-0.446	-1.343
Hedy scan	-0.788	0.261	0.561
Holb lati	1.077	-0.989	-0.453
Homa napa	-0.272	-0.813	0.515
Hoya lanc	-0.457	-0.639	0.290
Hydr febr	-0.821	-0.084	-1.207
Hydr sibt	-0.999	0.509	-0.905
Hype sp	0.294	-0.234	-0.566
Ilex aqui	2.905	0.816	-0.234
Inul capp	-0.766	0.946	0.856
Jasm offi	0.428	-0.532	-0.754
Jugl regi	-0.630	-0.588	-1.964
Koen moll	0.582	-1.261	-0.506
Lami albu	-0.999	0.509	-0.905
Ligu sine	0.086	-0.539	-0.553
Lind gran	-0.287	-0.376	-0.631
Lind sp	-0.391	-0.419	1.281
Lith eleg	-0.375	-0.793	-0.504
Lyon oval	-0.302	0.186	0.630

Mach odor	0.139	-0.808	-0.162
Madh long	-0.730	0.363	0.577
Maes chis	-0.470	-0.114	-1.342
Maho napa	-0.527	-0.458	-0.275
Mall phil	-0.456	-1.297	0.073
Mela mala	-1.002	-0.008	1.435
Moli capi	-0.613	0.767	-1.156
Muss macr	1.063	-0.477	0.075
Myri escu	0.006	-0.189	0.380
Myrs capi	-0.329	-0.750	0.708
Myrs segu	-0.955	-0.240	1.405
Myrs semi	0.155	-0.613	0.037
Neoc caud	0.759	-1.062	-1.965
Onyc japo	-0.935	1.009	-1.258
Onyc sili	-0.730	0.362	0.577
Ophi sp	-0.083	0.494	0.267
Opli burm	-0.472	0.139	0.999
Osbe nepa	-0.822	0.756	0.255
Osyw wigh	0.075	0.497	0.535
Oxys pani	-0.325	-0.239	-1.188
Part hyst	-0.999	0.509	-0.905
Pers duth	0.499	-0.696	-1.161
Pers chin	-0.651	-0.562	-1.362
Phyl niru	-0.817	0.677	0.961
Phyl reti	1.063	-0.477	0.075
Pile scri	-0.833	-0.446	-1.343
Pinu roxb	-0.970	0.542	0.143
Pinu wall	-0.488	1.241	1.213

Pipe betl	-0.418	0.160	0.596
Pipt nepa	2.905	0.816	-0.234
Poly poly	-0.061	0.055	0.252
Poly squa	-0.525	-0.882	0.199
Prim dent	2.905	0.816	-0.234
Prun cera	0.642	-0.986	-1.817
Prun sp	0.920	0.339	1.085
Prun spin	-0.132	-0.566	0.075
Pter aqui	-0.105	0.415	-0.403
Pter lati	-0.808	-0.244	-0.863
Pter vitt	-0.986	1.420	-1.459
Puer pedu	-0.488	1.241	1.213
Pyra cren	-0.688	0.500	-0.089
Pyru pash	0.084	-1.136	0.060
Quer flor	2.364	0.435	0.320
Quer glau	-0.099	0.538	0.696
Quer lame	-0.104	-0.050	0.472
Quer seme	0.789	1.036	0.383
Quer lana	0.420	-0.758	-0.091
Rand tetr	-1.463	0.098	1.136
Rhod arbo	0.475	-0.086	0.478
Rhus chin	-0.869	0.523	-0.565
Rhus java	1.449	-0.367	-0.108
Rhus wall	-0.433	-0.915	1.459
Rosa semp	-0.816	0.894	-0.676
Rosc purp	-0.369	-0.556	-0.994
Rubi manj	0.561	1.217	0.187
Rubu acum	0.551	-0.366	-0.320

Rubu elli	-0.301	0.988	-0.554
Rubu pani	0.311	-0.784	-0.509
Rubu ulmi	2.905	0.816	-0.234
Sarc hook	-0.664	0.131	-1.099
Saur napa	0.835	0.438	-0.081
Schi wall	-0.652	0.085	0.554
Sene wall	-0.065	-0.551	-1.284
Smil aspe	-0.237	0.066	-0.120
Smil eleg	1.242	-0.382	0.004
Smil fero	-0.763	-0.179	0.269
Smil glau	-0.025	-0.548	-0.964
Smil lanc	-0.524	-0.308	0.342
Smil oval	-1.463	0.098	1.135
Smil perf	-0.204	-1.297	-0.022
Smil zeyl	-0.401	-0.565	1.079
Sola virg	-0.861	-0.286	-1.270
Stac bull	0.168	0.067	-0.199
Step eleg	-0.595	-0.827	-0.661
Stro wall	-0.364	-0.004	-0.801
Syno capp	-0.287	-0.376	-0.631
Syzy cumi	-0.730	0.362	0.577
Tetr sp	0.306	-0.138	-0.396
Thal foli	1.544	-0.239	-1.014
Todd asia	-0.730	0.362	0.577
Urti dioi	-0.707	-0.438	-0.593
Vibu cyli	1.036	-0.701	0.465
Vite negu	-0.269	0.287	1.233
Zant arma	0.303	0.336	-0.580

b. Sites scores

Sites	CCA1	CCA2	CCA3
1	-0.956	1.156	-1.423
2	-0.821	-0.065	-0.733
3	-0.419	0.092	1.189
4	-0.123	0.894	0.419
5	-0.027	1.628	0.332
6	0.416	2.083	0.214
7	0.382	2.081	0.152
8	0.499	0.346	-0.020
9	-0.653	1.531	-1.320
10	-0.788	-0.574	1.054
11	-0.402	-1.209	0.401
12	-0.378	-1.357	0.454
13	4.701	1.813	-0.122
14	-1.124	-0.327	1.815
15	-0.656	-1.156	1.513
16	-0.056	-1.366	0.964
17	-0.052	-1.361	0.387
18	0.469	-1.225	-0.170
19	0.322	-0.750	-0.132
20	0.919	-0.758	-0.031
21	-0.913	-0.204	-2.282
22	-0.330	-1.008	-1.634
23	-0.188	-1.092	-0.502
24	-0.202	-0.543	0.861
25	-0.422	-0.118	1.019

26	0.200	-0.871	-1.353
27	0.896	-1.077	0.136
28	1.190	-1.616	-0.538
29	1.315	-0.634	-1.011
30	2.823	1.050	0.936
31	-1.169	0.148	2.678
32	-0.868	0.143	0.628
33	-0.619	-1.064	1.050
34	-0.145	0.507	0.812
35	-0.951	1.847	1.331