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Variation in Tree Density, Biomass and Carbon Stock with Altitude under the Large Cardamom-based Traditional Agroforestry System of Darjeeling Himalaya

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Abstract It has been reported that the large cardamom-based traditional agroforestry systems in the Darjeeling Himalaya of West Bengal, India represents a sustainable land use system with a high potential to store and sequester biomass carbon. Therefore, the present study was undertaken to evaluate the changes in tree biomass stock and carbon accumulation of this indigenous system with altitude. The study area was classified into three altitude-classes as low (700-1200 m asl), mid (1200-1700 m asl) and high (> 1700 m asl). The tree biomass and carbon storage for low-, mid- and high-altitude classes was estimated at 630.17 and 296.18 Mg ha⁻¹, 397.05 and 186.61 Mg ha⁻¹ and 315.78 and 148.42 Mg ha⁻¹, respectively. *Schima wallichii*, *Cryptomeria japonica* and *Cupressus cashmeriana* were the most ecologically important tree species based on IVI (important value index) in the low-, mid-, and high-altitude classes, respectively. The IVI and total biomass of the five most ecologically important tree species in low-, mid- and high-altitude ranged from 17.34-26.04, 18.15-37.56, 27.13-42.43 and 9.05-133.75 Mg ha⁻¹, 1.38-37.43 Mg ha⁻¹ and 19.0-72.1 Mg ha⁻¹, respectively. *Schima wallichii* occurred in all the altitude-classes among the five most ecologically important tree species. In the study area, the average ecosystem carbon storage was estimated at 295.02 Mg ha⁻¹. The contribution of soil organic carbon to the ecosystem carbon stock increased with the increasing altitude. In contrast, the contribution of biomass carbon to the ecosystem carbon stock decreased with the increase in altitude. The ecosystem carbon stock estimated for the low-altitude class was 36.43 % and 45.30 % higher than the mid- and high-altitude agroforestry systems. The large cardamom-based agroforestry systems in the Darjeeling Himalayas are thus a prospective carbon sink, both in vegetation and soil due to higher tree densities and natural resource conservation-based traditional farming practices.

Keyword Agroforestry system. Altitude. Biomass. Carbon storage. Ecosystem. Land use.

Introduction

The concern about global climate change is abundantly reported with a fear that it can potentially restrict the achievement of Millennium Development Goals (Jagermeyr, 2020; Tamang *et al.*, 2021; Pradhan *et al.*, 2022). The impacts of climate change are comparatively intense in the mountainous regions than in the plains (Palomo, 2017; Gioli *et al.*, 2019; Chhogyel *et al.*, 2020). The eastern Himalayan region is warming by 0.01-0.04 °C every year (Chaudhary *et al.*, 2011; Chettri *et al.*, 2012) with severe consequences on water resources, biodiversity, human health, agriculture, and consequently on food security throughout the region (Shukla *et al.*, 2016; Wangchuk and Wangdi, 2018; Vineeta *et al.*, 2023). One of the common land use systems of indigenous communities across the world is traditional agroforestry systems which are a viable option to increase land productivity while reducing global climate change impacts (Sharma *et al.*, 2016_a; Vineeta *et al.*, 2021, 2023). Diverse traditional agroforestry systems are practiced in the eastern Himalayas, which are lifeline for its inhabitants (Sharma *et al.*, 2016) and also are sustainable land management system that provides multiple ecosystem services (Prasad *et al.*, 2016; Feliciano *et al.*, 2018).

One such time-tested sustainable land management practice in Darjeeling Himalayas is large cardamom (*Amomum subulatum* Roxb.) based agroforestry system or under-canopy large cardamom cultivation, which has now attained research priority due to its associated ecosystem service benefits from provisional to regulative including economic benefits for farming households in the region (Sharma *et al.*, 2016_a; Vineeta *et al.*, 2021, 2022, 2023). Large cardamom, the oldest of spices, is native to Sikkim and Darjeeling Himalaya including, eastern hills of Nepal (Sharma *et al.*, 2016_{a-c}; Shrestha *et al.*, 2018). Its distribution is restricted mainly to Eastern Himalayan region of India, Nepal and Bhutan (Mehta *et al.*, 2015). It is a shade loving perennial herbaceous cash crop belonging to the family Zingiberaceae traditionally inter-mixed as an understorey of moist and semi evergreen natural forest trees on marginal lands and slopes with high moisture in areas of high rainfall between 1500-3500 mm at an altitude of 600 and 2000 m above mean sea level (Sharma *et al.*, 2009, 2016_{a-c}; Kumar *et al.*, 2015; Vineeta *et al.*, 2021, 2022, 2023). Of the many shade trees like *Alnus nepalensis*, *Schima wallichiana*, *Cryptomeria japonica* and *Cupressus cashmeriana* associated with the large cardamom, Nepalese or Himalayan Alder (*Alnus nepalensis*) is prominent because of its nitrogen fixing capacity (Negi *et al.*, 2018).

This traditional farming system is crucial for the region's local economy, contributing about 30 % of the total household income (Sharma *et al.*, 2016_b; Vineeta *et al.*, 2021, 2022, 2023). However, over the past decade, its area and productivity have declined significantly

73 due to climate change, insect pest, diseases and anthropogenic pressure (Sharma *et al.*, 2016a,
74 b; Shukla *et al.*, 2016). The cultivation area in Sikkim and Darjeeling Himalayas in 2011-12
75 was 23154 ha and 3305 ha, respectively, with production of 3237 tonnes and 626 tonnes,
76 respectively (Gudade *et al.*, 2013). In Darjeeling Himalayas during 2015-16 area of
77 cultivation, production and productivity of large cardamom was 2829 ha, 848.84 tonnes and
78 300.5 kg ha⁻¹; of which cultivation is prominently done in the district of Kalimpong (80 % of
79 the total cardamom area) (Tarafder *et al.*, 2018).

80 The altitudinal gradient significantly influenced trees' biomass and carbon stock
81 potential in agroforestry systems (Gebrewahid *et al.*, 2018). Biomass and carbon stock of
82 agroforestry systems in addition to vegetation, particularly woody species composition also
83 vary with locality factors, and geographic position or altitudes (Sharma *et al.*, 2016c;
84 Cardinael *et al.*, 2018; 2020; Feliciano *et al.*, 2018; Shi *et al.*, 2018; Corbeels *et al.*, 2019;
85 Yadav *et al.*, 2021). The main drivers of biomass and soil organic carbon (SOC) production,
86 incorporation, and mineralisation in an ecosystem are temperature and humidity (Tornquist *et al.*,
87 2009). With increasing altitude in Darjeeling Himalayas, annual precipitation increases
88 and mean temperatures decrease which reduces the decomposition of organic matter and
89 increases tree biomass production due to denser population and canopy which support higher
90 build-up of biomass carbon and SOC stock (Banerjee, 2014; Devi and Sherpa, 2019). SOC
91 was reported to increase with increasing altitude due to the cumulative annual addition of leaf
92 litter with slower decomposition rates (Dimri *et al.*, 1997; Banday *et al.*, 2019; Sheikh *et al.*,
93 2020). Increase in tree biomass and SOC with increasing elevation is due to increase in tree
94 density which produces denser canopy (Sheikh *et al.*, 2020) and more leaf litter while,
95 reducing temperature decreases decomposition rate inhibiting mineralization (Chan,
96 2008; Choudhury *et al.*, 2016).

97 The importance of tree-based systems in climate change mitigation and adaptation is
98 already brought to the forefront of various discussions and international forums (Zomer *et al.*,
99 2022). The biomass storage in this tree-based system in the Sikkim Himalayan regions have
100 been reported by Singh *et al.* (2018) and Lepcha and Devi (2020). However, the biomass
101 carbon storage of this system along an altitudinal gradient in the Darjeeling Himalayas has
102 not been reported. The large cardamom-based agroforestry systems in the Darjeeling
103 Himalayas are highly peculiar due to their diverse structure and functionality (Singh *et al.*,
104 2018). They are often regarded as the shadow of natural forests and offer multiple benefits
105 from ecological to livelihood, including restricting carbon emissions and ensuring socio-
106 economic security (Yadav *et al.*, 2021). Academic research studies still need to pay attention

to this traditional farming system of the region in terms of distribution and relationship between on-system tree biomass and carbon stock along the altitudinal gradient (Sharma *et al.*, 2016c; Singh *et al.*, 2018). Data on natural resources of Himalayan region is deficient and less studied due to poor accessibility, infrastructure and high spatio-temporal variability (Pandey *et al.*, 2017; Vineeta *et al.*, 2023). In this context, large cardamom-based agroforestry systems' biomass and carbon storage potential need to be evaluated.

Studies on large cardamom-based traditional agroforestry system in respect to biomass production, SOC and carbon storage capacity were very few mainly reported from Sikkim Himalayas (Sharma *et al.*, 2009; 2016a-c) but none compared the large cardamom-based traditional agroforestry system on aspect of altitudinal gradient. This traditional farming system of the region has still been ignored by academic research studies in terms of distribution and relationship between on system tree biomass, and carbon stock along the altitudinal gradient (Sharma *et al.*, 2016c; Singh *et al.*, 2018). Data on natural resources of Himalayan region is deficient and less studied due to poor accessibility, infrastructure and high spatio-temporal variability (Pandey *et al.*, 2017; Vineeta *et al.*, 2023).

A few studies from Darjeeling Himalayas evaluated vegetation diversity pattern along the elevation gradient (Das and Ghosh, 2022; Vineeta *et al.*, 2022) while, a few studies also quantified biomass and carbon stock (Banerjee and Prakasham, 2012; Gangopadhyay *et al.*, 2021) but none of these studies examined the effect of altitude on biomass and carbon stock. Therefore, in continuation of our earlier study (Vineeta *et al.*, 2022) where we examined the relationship of species richness, plant population and diversity of the traditional large cardamom-based agroforestry systems in Darjeeling Himalayas with altitude, the present study was executed in the same systems with following specific research questions:

- Does any relationship exist between altitude and tree density, biomass, biomass carbon and SOC stock under the traditional large cardamom-based agroforestry system in Darjeeling Himalayas?
- What are the prominent tree species in this system along the altitudinal gradient?
- Does tree density of the system influence its biomass, biomass carbon and SOC?

With reference to the earlier studies on the effect of elevational gradient on plant biomass, biomass carbon and SOC stock of the forest types in the Darjeeling Himalayas (Banerjee, 2014; Devi and Sherpa, 2019), we hypothesised that tree density, biomass and biomass carbon and SOC stock of traditional large cardamom-based agroforestry system in Darjeeling Himalayas will also increase with increasing elevation and with higher tree density the tree biomass carbon and SOC will also be higher. With the above-mentioned

specific research questions and hypothesis, we attempted to generate precise and systematic quantitative data of large cardamom-based traditional agroforestry systems in the Darjeeling Himalayas along the altitudinal gradient with the following specific objectives:

- To evaluate the variation in tree density of the system across the altitudinal gradient.
- To evaluate the biomass and biomass carbon storage of the system across the altitudinal gradient.
- To evaluate the soil organic carbon storage of the system across the altitudinal gradient.
- To evaluate the influence of tree density on the systems tree biomass, biomass carbon and SOC stock.

Material and Methods

Site Description

The present study was conducted in the Darjeeling Himalayan region of West Bengal, India (Fig. 1) from, January 2019 to April 2021. The study area includes two administrative districts of Kalimpong and Darjeeling but exclusive the plain or Terai region of Darjeeling district i.e., Siliguri sub-division. The study site extends between 26° 27' 05"-27° 13' 10" N latitude and 87° 59' 30"-88° 53' E longitude with an altitude of 132-3660 m. The region is humid and sub-tropical to sub-alpine where the average temperature varies between 3 and 22°C (Saxena, 2005). The average annual rainfall is between 270-310 cm and the soils of the region are acidic, yellow to red-brown in color and silty loam to sandy loam textured (Froehlich and Sarkar, 2000). Tropical (below 800 m), subtropical (800-1600 m), temperate (1600-2400 m), cold-temperate (2400-3200 m) and sub-alpine (3200-4000 m) types of vegetation are found in the Darjeeling Himalayas (Cajee, 2018). The forests in the region are mostly reserved and protected. Forests (33957 ha) and agriculture including tea gardens (131063 ha) are the prominent land uses of Darjeeling Himalayas (www.darjeeling.gov.in).

Sampling Design

Before starting our research, we conducted a thorough reconnaissance survey of the study area to explore the large cardamom-based traditional agroforestry systems. Traditionally large cardamom-based agroforestry farming in Darjeeling and Sikkim Himalayas were reported as semi-domesticated systems where large cardamom is cultivated under the canopy of reserved or protected forest leased out to the growers by the State Forest Department with no rights to cut the trees (Sharma *et al.*, 1994, 2009). We could locate only 25 traditional large cardamom-based agroforestry systems/holdings in the study area which were about 1-3 ha in area in the elevations between 700-2000 m, of which 17 were in Kalimpong district and only eight were in Darjeeling district. We didn't consider the large cardamom under-canopy cultivation in agricultural landscapes for the present study because

these cultivations were adopted not more than a decade ago, that too under the canopy of planted trees. We thus selected all the 25 traditional large cardamom-based agroforestry holdings purposively for the study. Following the altitudinal chronosequence for the distribution of vegetation in the Darjeeling Himalayas (Cajee, 2018), we classified the large cardamom-based agroforestry holdings according to their altitudinal locations as low (700-1200 m asl), mid (1200-1700 m asl) and high (> 1700 m asl) with eleven, nine and five holdings, respectively. The geographical coordinates of the holdings were recorded with Garmin 72.

We adopted a stratified random nested quadrat sampling method laying 1-4 quadrates of size 20 m x 20 m (Vineeta *et al.*, 2022). Holdings with one hectare were laid with only quadrat while, 2-4 quadrates were marked diagonally across the corners.

Estimation of tree density

Abundance of a species in a unit area is expressed as density (D) which indicates the numerical strength of a species in its community.

Estimation of importance value index

Importance value index (IVI) reflects the sociological structure of a species in a community as it indicates its importance in the community. It is summation of relative frequency (RF), relative density (RD) and relative abundance (RA) of a species are its IVI (Kershaw, 1973; Cintron and Novelli, 1984; Brower *et al.*, 1997). We selected the prominent tree species in the different altitudinal classes based on their IVI values.

RD is per cent representation of a species in term of number of individuals relative to all other species in a community.

$$RD = \frac{\text{No. of individuals of the species}}{\text{No. of individuals of all species}} \times 100$$

Abundance of a particular species relative to total number of individuals of all species in the sampled area is RA.

$$RA = \frac{\text{Abundance of a species}}{\text{Sum of the abundance of all species}} \times 100$$

The frequency of a species relative to frequency of all other species in a community is RF and can be determined by using the following formula:

$$RF = \frac{\text{No. of occurrence of a species}}{\text{No. of occurrence of all species}} \times 100$$

Estimation of biomass carbon

Indirect or non-destructive method was adopted for estimating the above ground biomass (AGB) of trees following an allometric model suggested by Nath *et al.* (2019) because law of the land doesn't permit destructive sampling in the forest lands.

$AGB = 0.18D^{2.16} \times 1.32$; where D is diameter at breast height measured by tree caliper.

The below ground biomass (BGB) was estimated by multiplying the AGB by a factor of 0.25 as per IPCC guideline (2006) and Huang *et al.* (2020). The total trees biomass obtained by the summation of AGB and BGB and the tree biomass carbon was estimated as 47% of the biomass in the tree (IPCC, 2006).

Estimation of SOC

Soil samples were collected from three points (two at the corners and one at centre) laid out diagonally in a quadrat. Composite soil samples from every quadrat were collected at 0-20, 20-40 and 40-60 cm soil depths separately. Samples were air dried in shade, ground with wooden pestle, passed through 2 mm sieve and stored in cloth bags for further laboratory analysis. Walkley and Black's rapid titration method was adopted for estimating the organic carbon content in the soil (Jackson, 1967) and the carbon stock in the soil was calculated by multiplying the organic carbon with the weight of the soil (depth x bulk density) for a particular depth and expressed in megagrams (Mg) per ha (Joao Carlos *et al.*, 2001). We adopted Core sampler method to estimate bulk density (Piper, 1950).

Statistical tools

The data were subjected to analytical statistical tools like analysis of variance, Karl Pearson's coefficient of correlation and regressions (Gomez and Gomez, 1984; Goldstein and Dillon, 1985) using IBM SPSS version 2020.

Results and Discussion

Tree density and diameter

Tree density and diameter at breast height (dbh) among the altitude-classes didn't vary significantly (Table 1).

Table 1. Tree density, biomass (Mg ha⁻¹), and carbon of large cardamom based traditional agroforestry systems at different altitude classes

AC	D	Dy	AGB	BGB	TB	AGC	BGC	TBC	SOC	ES
Low	25.4	88	504.14	126.03	630.17	236.94	59.24	296.18	66.92	363.10
Medium	43.2	125	317.64	79.41	397.05	149.29	37.32	186.61	85.45	272.06

High	45.9	104	252.63	63.16	315.78	118.73	29.68	148.42	101.48	249.90
LSD_{0.05}	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Overall	38.2	106	358.14	89.53	447.67	168.32	42.08	210.40	84.62	295.02

AC- Altitude class (Low- 700-1200 m asl; Medium- 1200-1700 m asl; High- > 1700 m asl); D- Diameter at breast height (cm); Dy- Density- (trees ha⁻¹); AGB- Above ground biomass (Mg ha⁻¹); BGB- Below ground biomass (Mg ha⁻¹); TB- Tree biomass (Mg ha⁻¹); AGC- Above ground carbon (Mg ha⁻¹); BGC- Below ground carbon (Mg ha⁻¹); TBC- Tree biomass carbon (Mg ha⁻¹); ES- Ecosystem carbon (Mg ha⁻¹)

However, tree density increased by 29.6 % from low- to medium-altitude class while, it decreased by 16.8 % from mid- to high-altitude class and tree density of high-altitude class was 18.18 % more than the low-altitude class. Similarly, tree dbh gradually increased from one altitudinal class to another with 70.1 % and 6.2 % increase from low to medium and medium to high-altitudinal class, respectively while, dbh of high-altitude class was 80.7 % more than dbh of low altitude class. Overall, the tree density and dbh estimated for the system ranged from 5.0-210.0 trees ha⁻¹ and 8.0-54.8 cm, respectively (Supplementary table 1). *Litsea glutinosa* was estimated with lowest density and top five tree species based on density in the system were *Cryptomeria japonica* (210.0 trees ha⁻¹), *Schima wallichii* (205 trees ha⁻¹), *Ficus auriculata* (190 trees ha⁻¹), *Alnus nepalensis* (155 trees ha⁻¹), and *Litsea monopetala* (130 trees ha⁻¹) while, *Leucosceptrum canum* was measured with smallest dbh and top five tree species based on dbh were *Cryptomeria japonica* (54.8 cm), *Alnus nepalensis* (39.5 cm), *Schima wallichii* (38.2 cm), *Cupressus cashmeriana* (33.1 cm), and *Toona ciliata* (31.8 cm).

Tree density estimated at low-, medium- and high-altitude class was in the range of 30-205 trees ha⁻¹ (*Cryptomeria japonica*-*Schima wallichii*), 20-210 trees ha⁻¹ (*Exbucklandia populnea*-*Cryptomeria*), and 5-145 trees ha⁻¹ (*Litsea glutinosa*- *Cupressus cashmeriana*), respectively. *Ficus auriculata* (190 trees ha⁻¹), *Litsea monopetala* (130 trees ha⁻¹), *Firmiana colorata* (120 trees ha⁻¹), and *Ficus colorata* (115 trees ha⁻¹) were the other denser tree species found at low-altitude class. In the mid-altitude, other denser trees were *Alnus nepalensis* (155 trees ha⁻¹), *Schima wallichii* (95 trees ha⁻¹), *Ficus semicordata* (85 trees ha⁻¹), and *Brassaiopsis mitis* (80 trees ha⁻¹). At high altitude class, the other denser species found were *Alnus nepalensis* (120 trees ha⁻¹), *Cryptomeria japonica* (110 trees ha⁻¹), *Euonymus attenuatus* (80 trees ha⁻¹), and *Schima wallichii* (75 trees ha⁻¹). Similarly, tree dbh at low-, medium- and high-altitude class was measured in the range of 12.7-54.8 cm (*Citrus spp.*-*Cryptomeria japonica*), 8.0-32.5 cm (*Leucosceptrum canum*-*Cryptomeria japonica*), and 14.0-38.2 cm (*Pinus wallichiana*-*Schima wallichii*), respectively. Other bigger trees in terms of dbh in low-altitude class found were *Alnus nepalensis* (39.5 cm), *Schima wallichii* (33.7

cm), *Erythrina variegata* (31.2 cm), and *Ficus semicordata* (29.9 cm), in mid-altitude class were *Toona ciliata* (31.8 cm), *Alnus nepalensis* (29.9 cm), *Schima wallichii* (28.7 cm), and *Macaranga denticulata* (28.0 cm) and in high-altitude class were *Cupressus cashmeriana* (33.1 cm), *Cryptomeria japonica* (28.7 cm), *Euonymus attenuatus* (28.0 cm), and *Alnus nepalensis* (27.1 cm).

In contrast to the Sikkim Himalayan large cardamom based traditional agroforestry systems, where *Alnus nepalensis* reported as the prominent shade tree with density of 80.13 tree ha⁻¹ (Lepcha and Devi, 2020), the prominent trees found in the Darjeeling Himalayan systems were *Schima wallichii* (205.0 trees ha⁻¹), *Cryptomeria japonica* (210 trees ha⁻¹) and *Cupressus cashmeriana* (145 trees ha⁻¹) at low-, mid- and high-altitude class respectively. In this study *Alnus nepalensis* was found with density of only 35 trees ha⁻¹ at low altitude class while, at mid- and high-altitude class the species followed *Cryptomeria japonica* and *Cupressus cashmeriana* with density of 155 and 120 trees ha⁻¹, respectively. However, the density of *Alnus nepalensis* estimated at mid- and high-altitude classes in the present study is 39.87-74.87 trees ha⁻¹ than estimated at Sikkim Himalayas (80.13 trees ha⁻¹) in the altitude of 1350 to 1619 m above mean sea level. This was because the studied systems at Sikkim Himalayas were in the agricultural landscape planted 20 years ago where the growers preferred *Alnus nepalensis* to maintain soil fertility and productivity as it can fix atmospheric nitrogen (Sharma *et al.*, 1994). In contrast, the Darjeeling systems were cultivated under-canopy of leased out forest land by the growers with no rights to change the natural species composition of the forest. However, total tree density estimated in this study (105.33 trees ha⁻¹) was lesser than the Sikkim Himalayan studies (124.19 trees ha⁻¹) because the Darjeeling Himalayan systems were forest-based (Sharma *et al.*, 2007) where the dominance of the species was distributed as compared to the Sikkim Himalayan system which was planted at an agricultural landscape where preferred species were planted and allowed to grow.

The tree density of the Darjeeling Himalayan system was also either comparable or higher than the poplar agroforestry system of North-western India (Rizvi *et al.*, 2011), jhum fallow agroforestry of Tripura (Chaudhary *et al.*, 2016), homegardens of Terai region (Subba *et al.*, 2017, 2018) and Darjeeling Himalayas (Sarkar, 2020) of West Bengal, poplar agroforestry systems of China (Fang *et al.*, 2010) and coffee agroforestry of Guatemala (Schmitt-Harsh *et al.*, 2012). This indicates that the under-canopy forest-based traditional large cardamom agroforestry systems have the potential to promote, harbour and maintain diverse tree species with higher overall population due to their retained structure and

composition as of the original forest. This also results in higher tree growth and thus acts in a way very similar to a natural forest (Sharma *et al.*, 2007). However, the tree diameter of the systems was found lesser than the Sikkim systems (Lepcha and Devi, 2020), tea agroforestry of Assam (Kalita *et al.*, 2016), managed plantation and jhum fallow agroforestry of Tripura (Chaudhary *et al.*, 2016) and homegardens of terai region (Subba *et al.*, 2017, 2018) and Darjeeling Himalayas (Sarkar, 2020) of West Bengal. This might be due to the predominance of trees of young age and the higher heterogeneity of the Darjeeling systems in terms of its structure and composition where dominance by the species was determined by higher inter- and intra-specific competition (Odum, 1971, 1983).

Tree biomass and carbon stock

The biomass and carbon stock estimated for low-, mid- and high-altitude classes of large cardamom-based traditional agroforestry systems of Darjeeling Himalaya is presented in Table 1 and Supplementary table 1. The tree biomass and carbon decreased gradually with the increasing altitude classes but the changes were non-significant. Tree biomass stock decreased by 36.99 % from low- to mid-altitude, 20.47 % from mid- to high-altitude and 49.89 % from low- to high-altitude classes due to variation in tree dbh, density and species richness (Supplementary table 2; Vineeta *et al.*, 2022). The contribution of the above ground biomass (AGB) was 80 % at all the altitude classes. The changes in tree biomass storage along the altitude classes were inversely related ($r = -0.255^*$). A non-significant effect of altitude on forest biomass and carbon was earlier reported (Mensah *et al.*, 2016; Wondimu *et al.*, 2021), while, on the contrary, either direct or inverse relationships between elevation and biomass or carbon stock also exist (Ensslin *et al.*, 2015; Zhang *et al.*, 2018). Varied relationship on biomass and biomass carbon attributed to varied range of altitude with variation in tree species richness, population, and their composition (Wondimu *et al.*, 2021).

The large cardamom-based traditional agroforestry systems at low altitude-class accumulated highest tree biomass and carbon in spite of lowest tree density and diameter because of a greater number of species having density higher than 100 trees ha⁻¹ and dbh of 25 cm as compare to mid- and high-altitude class (Supplementary table 2). This was also evidenced from linear regression models between tree density and tree diameter ($r^2 = 0.05863$, $P < 0.05$; fig. 56) and tree biomass density and tree diameter ($r^2 = 0.407$, $P < 0.01$; Fig. 2). Similar, relationship between tree biomass/tree biomass carbon and tree density was also reported from large cardamom-based traditional agroforestry systems from Sikkim Himalayas (Lepcha and Devi, 2020), Darjeeling Himalayan homegardens (Sarkar, 2020) and for different subtropical ecosystems of Northeast India (Borah *et al.*, 2015; Chaudhary *et al.*,

2016). In our earlier study, we have reported a reverse and significant relationship of species richness of this large cardamom-based traditional agroforestry system with altitude (Vineeta *et al.*, 2022). Higher tree species richness in low-altitude class than mid- and high-altitude class might also be responsible for higher tree biomass in low-altitude class (Supplementary table 1). Significant and positive influence of species richness on forest biomass and biomass across the altitudinal gradient was reported by many studies (Mensah *et al.*, 2016; 2017; Wondimu *et al.*, 2021). Direct relationship of biomass or carbon accumulation with species richness was attributed to various biotic interactions like facilitation e.g., improving soil fertility by litter fall which explains land uses with heterogeneous vegetation has higher productivity than the land uses with homogenous vegetation (Rai *et al.*, 2021; Wondimu *et al.*, 2021).

Total tree biomass and carbon storage quantified in this study is higher than the Sikkim Himalayan systems (Lepcha and Devi, 2020), homegardens of the Terai zone (Subba *et al.*, 2017, 2018) and Darjeeling Himalayas (Sarkar, 2020) of West Bengal and other agroforestry systems elsewhere (Kalita *et al.*, 2016; Yadav *et al.*, 2017). However, it is comparable to different subtropical forest ecosystems of Indian eastern Himalayas (Borah *et al.*, 2015; Chaudhary *et al.*, 2016). The variation in biomass and carbon accumulation amongst these studies is due to variation in geographical location (site quality, topography, climate and edaphic conditions), plant species and species diversity (stem density, stem size distribution, age, density, diameter, canopy height and wood density), management practices and disturbance history (Liu *et al.*, 2015; Kalita *et al.*, 2016; Devi and Yadava, 2016; Rai *et al.*, 2021; Tamang *et al.*, 2021).

IVI and total biomass of dominant species

The IVI value indicates the ecological significance of a species in a particular area, accordingly, species having a higher IVI value have higher relative dominance than other species in the study area (Zafriakma *et al.*, 2020). In the present study, five species with top IVI values were selected from each of the altitudinal classes (Supplementary table 1) and evaluated their potential contribution toward total biomass (Fig. 3-8). *Schima wallichii*, *Cryptomeria japonica*, and *Cupressus cashmeriana* were the dominant species reported from the low-, mid-, and high altitudinal classes respectively. In addition, *Schima wallichii* was one of the top five dominant species in all the altitudinal classes. It is also evident that the total biomass carbon was higher for the species with higher IVI in each of the altitude classes. While comparing the total biomass of the species with higher IVI among the three altitudinal classes, maximum biomass was observed with the species in the lowest altitudinal class

(*Schima wallichii*, 133.75 Mg ha⁻¹) followed by the high- (*Cupressus cashmeriana*, 72.1 Mg ha⁻¹) and mid-altitudinal classes (*Cryptomeria japonica* (37.43 Mg ha⁻¹)).

The potential biomass and carbon storage varied from species to species and also with altitude class due to variations in tree density and dbh (Table 2). At the low-altitude class, the most prominent tree species in terms of biomass accumulation was *Schima wallichii* with an estimated biomass of 121.8 Mg ha⁻¹ and average DBH and density of 33.7 cm and 205 trees ha⁻¹ respectively. It was followed by *Cryptomeria japonica* (estimated biomass of 50.7 Mg ha⁻¹, dbh 54.8 cm and density 30 trees ha⁻¹) and *Ficus semicordata* (biomass of 50.4 Mg ha⁻¹, dbh 29.9 cm and density 110 trees ha⁻¹). In the mid-altitudinal class, the major tree found was *Cryptomeria japonica* with biomass of 114.9 Mg ha⁻¹, dbh 32.5 cm and density of 210 trees ha⁻¹, followed by *Alnus nepalensis* (biomass of 71.2 Mg ha⁻¹ with dbh 29.9 cm and density 155 trees ha⁻¹) and *Schima wallichii* (biomass of 39.6 Mg ha⁻¹ with dbh 28.7 cm and density 95 trees ha⁻¹). *Cupressus cashmeriana*, *Schima wallichii*, *Cryptomeria japonica* and *Alnus nepalensis* were the tree species with higher IVI found at high-altitude class with biomass accumulation of 82.7, 58.3, 45.9 and 44.3 Mg ha⁻¹ with a dbh of 33.1, 38.2, 28.7 and 27.1 cm and density of 145, 75, 110 and 120 trees ha⁻¹, respectively.

In contrast to maximum biomass and carbon stock by *Alnus nepalensis* at Sikkim Himalayan systems, *Schima wallichii*, *Cryptomeria japonica* and *Cupressus cashmeriana* reported the maximum biomass and carbon at low-, middle- and high-altitude classes respectively. However, similar to the Sikkim systems, the highest biomass and carbon accumulation in this study was also found to correspond with the highest tree density and dbh (Lepcha and Devi, 2020). The tree biomass and carbon storage estimated for different species in the present study were found inconsistent with the dbh of the trees but largely consistent with the tree density, similar to the studies reported for large cardamom systems of Sikkim Himalayas (Lepcha and Devi, 2020), and tea agroforestry system of Assam (Kalita *et al.*, 2016) while, contrasted to *Dipterocarpus* forest of Manipur (Devi and Yadava, 2016). Similarly, above and below-ground biomass allocation of many individual tree species was reported with a major contribution by above-ground biomass (Justine *et al.*, 2015; Singh and Singh, 2017). Large cardamom based traditional agroforestry systems in the present study exhibited many shade-tree including *Alnus nepalensis* which provide multiple ecosystem services from provisions like fuelwood and fodder to regulatory like maintaining soil fertility and productivity (Lepcha and Devi, 2020; Cyamweshi *et al.*, 2021). *Alnus nepalensis* was reported as dominant shade tree species in large cardamom based traditional agroforestry

systems of Sikkim Himalayas (Sharma *et al.*, 2009; Singh *et al.*, 2018; Lepcha and Devi, 2020) but in present study the species was found as one of the many species present as shade-trees in the large-cardamom systems.

The estimation of shade-tree biomass stock and its allocation will aid us with better insight of life history strategies of these trees like nutrient cycling, carbon stocks, and fuel property including their potential to offset carbon dioxide emission (Pickup *et al.*, 2005; Niinemets *et al.*, 2007) to develop sustainable land management options for the Darjeeling Himalayas with these traditional large cardamom-based agroforestry systems (Nair and Nair, 2014; Possu *et al.*, 2018; Shukla and Chakravarty, 2018). Understanding the quantum of biomass accumulation and allocation of the shade-trees of the large cardamom-based traditional agroforestry systems indicated the productivity of ecologically most important tree species in the system including their and the system's contribution towards net carbon emission and their potential to sequester carbon (Chhabra and Dadhwal, 2004). These tree species in the system are crucial as they regulate the energy flow and material cycles (Devi and Yadava, 2009). These large cardamom-based traditional agroforestry systems are sequestering carbon permanently because the system is forest based that too on reserved forests where the shade-tree species will never ever be removed like other agroforestry systems (Chakravarty *et al.*, 2017_{a, b}; Mengistu and Asfaw 2019; Vineeta *et al.*, 2021).

Soil organic carbon stock

Oxidizable soil organic carbon (SOC) carbon stock up to 60 cm soil depth increased gradually with increasing altitude but the changes were statistically similar (Table 1). The SOC stock increased by 27.69 %, 18.76 % and 51.64 % from low- to mid-, mid- to high- and low- to high-altitude class, respectively. Overall, SOC stock up to 60 cm soil depth quantified was 84.62 Mg ha⁻¹ in the range of 66.92-101.48 Mg ha⁻¹ in the present study is within the range of 8.9 to 851.9 Mg C ha⁻¹ estimated for agroforestry systems, plantations, and forests (Shukla and Chakravarty, 2018; Shukla *et al.*, 2018; Simon *et al.*, 2018; 2020; Rai *et al.*, 2021; Chettri *et al.*, 2023; Dey *et al.*, 2023). However, SOC stock significantly increased with increasing altitude ($r = 0.699^{**}$) due to slower decomposition rates similarly reported from Sikkimese large cardamom agroforestry systems (Lepcha and Devi, 2020), homegardens of Darjeeling Himalayas (Sarkar, 2020) and other different land-uses elsewhere (Ramesh *et al.*, 2015; Chaudhary *et al.*, 2016). Lower temperature at higher altitude increases soil acidity and anaerobic conditions which inhibit the decomposition and mineralization process cumulating the build-up of undecomposed organic matter (Devi and Sherpa, 2019; Lepcha and Devi, 2020; Choudhury *et al.*, 2021).

Distribution and composition of vegetation is greatly influence by altitude as temperature decreases with increasing altitude which reduces the diversity and population thus regulating carbon (Shedayi *et al.*, 2016) as altitude was found prime determinant of ecosystem properties and processes in the mountainous regions (He *et al.*, 2016). In our earlier study involving large cardamom-based traditional agroforestry systems of Darjeeling Himalayas we also found diversity and plant population reduced as altitude increased (Vineeta *et al.*, 2022). Additionally, SOC is also influenced by soil structure, species composition, organic matter input (quality and quantity), locality factors (climatic, soil and topographic variables), land use and management (Shukla and Chakravarty, 2012; Simon *et al.*, 2018; Devi and Sherpa, 2019).). In Darjeeling Himalayas, the plant assemblages of traditional below-canopy forest-based large cardamom agroforestry systems are highly diverse (Vineeta *et al.*, 2022) where organic input is contributed by the entire system vegetation building-up higher SOC than the agroforestry systems at agricultural landscapes where only trees contribute to SOC build-up (Bishaw *et al.*, 2013; Choudhury *et al.*, 2021). The influence of altitude is a priority area of research to study ecosystem and climate change (Malhi *et al.*, 2010). Traditional under-canopy forest-based large cardamom agroforestry systems can be a climate change mitigation option in Darjeeling Himalayas due to their soil carbon sequestration potential to increase carbon footprint for land use productivity and sustainability. This is because tree-based land use systems were always reported with higher soil carbon content than any other land uses with possibility of higher net gains in carbon stocks (Minasny *et al.*, 2017; Simon *et al.* 2018; Lepcha and Devi, 2020; Panwar *et al.*, 2022).

Ecosystem carbon stock

The ecosystem carbon stock gradually decreased as altitude increased but didn't vary significantly among the three altitude-classes (Table 1). The ecosystem carbon stock decreased by 25.07 %, 8.14 % and 31.17 % from low- to mid-, mid- to high- and low- to high-altitude class, respectively. Overall, it was estimated at 295.02 Mg ha⁻¹ with 71.32 % and 28.68 % contribution by tree biomass and soil, respectively. The contribution of SOC increased and tree biomass carbon decreased to ecosystem carbon stock with increasing altitude.

Ecosystem carbon stock under large cardamom traditional agroforestry systems estimated was higher than the stock estimated for traditional homegardens, farm forestry and plantation forests (Mulatu, 2019; Tamang *et al.*, 2021; Chettri *et al.*, 2023; Dey *et al.*, 2023). The carbon sequestration potential of tree-based land use systems varies with geographical

location, nature of species, species richness and age of the tree (Liu *et al.*, 2015). Moreover, the amount of carbon stored in large cardamom based traditional agroforestry system was reported to be predominantly dependent on its structure and function, which in turn were influenced by the community and culture of the region (Singh *et al.*, 2018; Lepcha and Devi, 2020). Higher biomass and SOC stock can efficiently and permanently manage the ecosystem carbon pool in large cardamom-based traditional agroforestry systems (Mathew *et al.*, 2016). The quantified amount of ecosystem carbon stock of the studied large cardamom-based agroforestry systems of Darjeeling Himalayas indicate that the system is a prospective carbon sink both in vegetation and soil due to its higher tree density and natural resource conservation-based traditional farming practices. Large cardamom-based agroforestry systems are practised in leased-out reserved forests areas where tree felling is prohibited. Consequently, the vegetation heterogeneity of these under-canopy forest-based large cardamom traditional agroforestry systems is conserved which leads to higher carbon net gain and high ecosystem carbon balance while, making the system more permanent, stable, resilient and productive than any other land use systems in an agricultural landscape (Mulatu, 2019; Rai *et al.*, 2021; Tamang *et al.*, 2021; Chettri *et al.*, 2023; Dey *et al.*, 2023). This traditional practice of forest-based under-canopy large cardamom cultivation can be an efficient adaptation option at Darjeeling Himalayas to mitigate climate change locally and regionally besides the provision of livelihood and ecological benefits to the locals (Lepcha and Devi, 2020; Vineeta *et al.*, 2023). Managing soil and biomass carbon in this traditional system will both be an avoided emission and net addition of carbon to terrestrial pool thereby fulfilling the global 4 per mille targets in conformity of the Kyoto Protocol under article 3.3 (Lal, 2016; Minasny *et al.*, 2017).

Conclusion

The traditional large cardamom farming systems are diverse, structurally and compositionally, thus making the system resilient and productive. Therefore, identifying the ecologically most important tree species based on IVI will help sustainable management of the system since these species mainly regulate the magnitude and pattern of energy flow and material cycle between biotic and abiotic components of the system. The density of trees in the system was in the range of 5.0-210.0 trees ha⁻¹ with *Schima wallichii* (205 trees ha⁻¹), *Ficus auriculata* (190 trees ha⁻¹) and *Alnus nepalensis* (155 trees ha⁻¹) as prominent tree species. In addition, biomass carbon data of different altitude-classes will enhance our understanding of the life history, nutrient cycling, carbon stocks, and potential to offset carbon dioxide emissions of the system. The tree biomass storage gradually decreased with

the increasing altitude-classes as was evidenced from significant inverse relationship between tree biomass accumulation and altitude. The tree biomass and ecosystem carbon accumulated in the system quantified was 447.67 and 295.02 Mg ha⁻¹ while, at low-, mid- and high-altitude class the biomass estimated was 630.17 and 363.1 Mg ha⁻¹, 397.05 and 272.06 Mg ha⁻¹ and 315.78 and 249.09 Mg ha⁻¹, respectively. *Schima wallichii*, *Cryptomeria japonica*, and *Cupressus cashmeriana* were the dominant. Moreover, species in terms of their highest IVI values in low-, mid-, and high-altitude classes, respectively. The ecologically most important tree species in each altitude classes were also quantified with higher total biomass carbon. The high ecosystem carbon storage under the forest-based traditional large cardamom agroforestry systems indicates that management and promotion of such systems can be an efficient adaptation option in Darjeeling Himalayas to mitigate climate change locally and regionally in addition to the provision of livelihood and other ecological benefits.

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Declarations

Conflict of interest: The authors declare that they have no conflict of interests.

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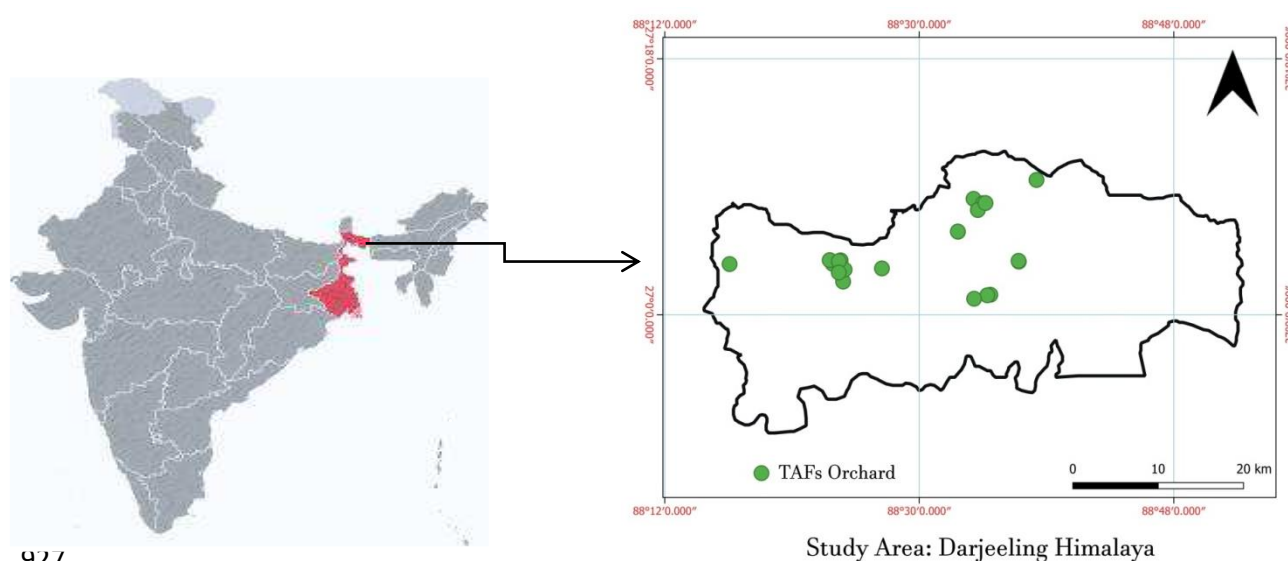


Fig. 1. Outline map of study area

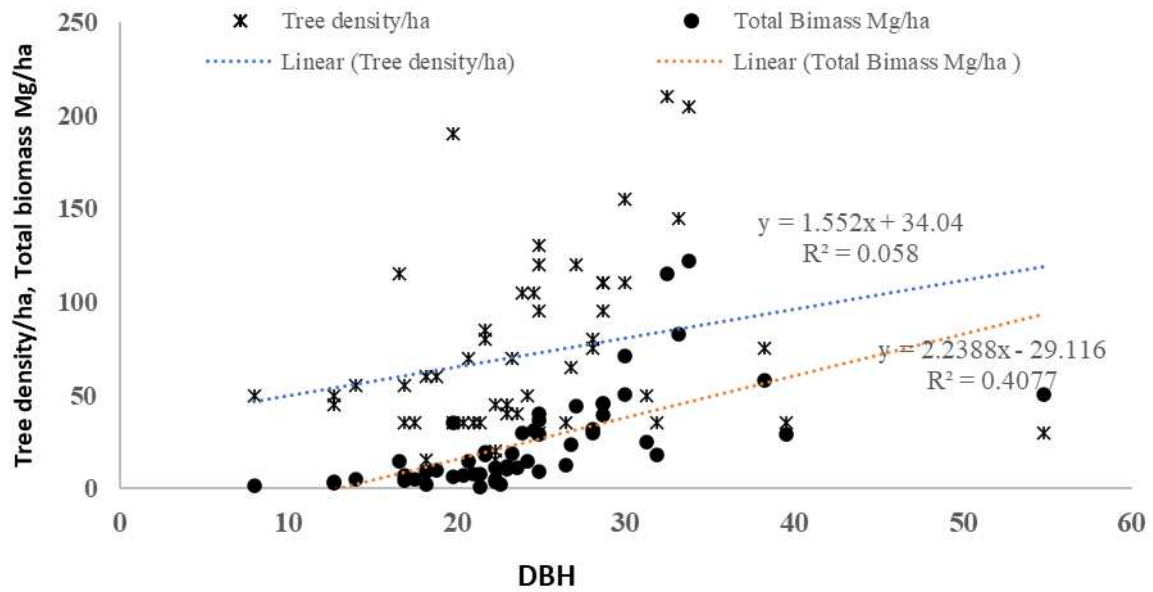


Fig. 2. Regression models tree density and tree diameter and tree biomass density and tree diameter

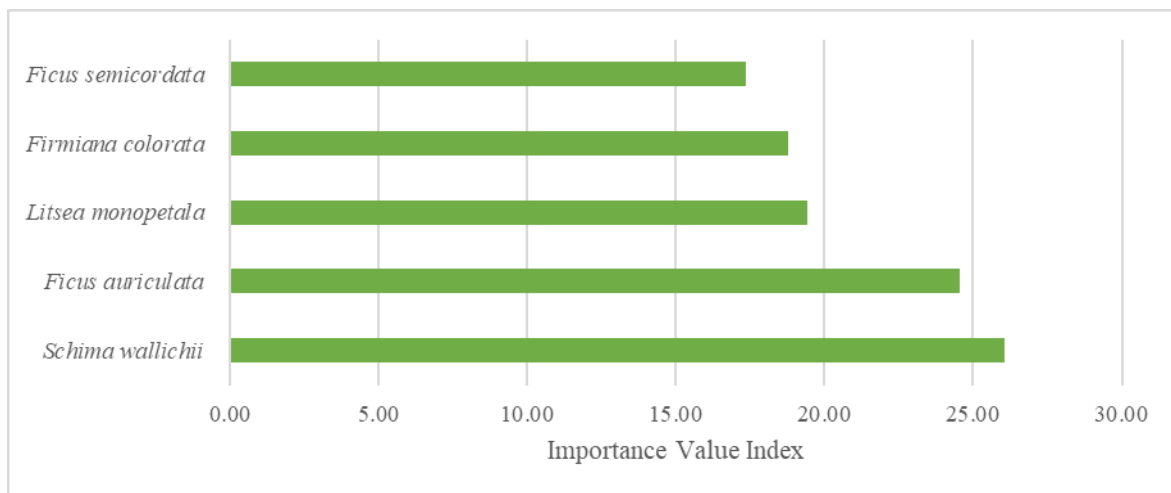
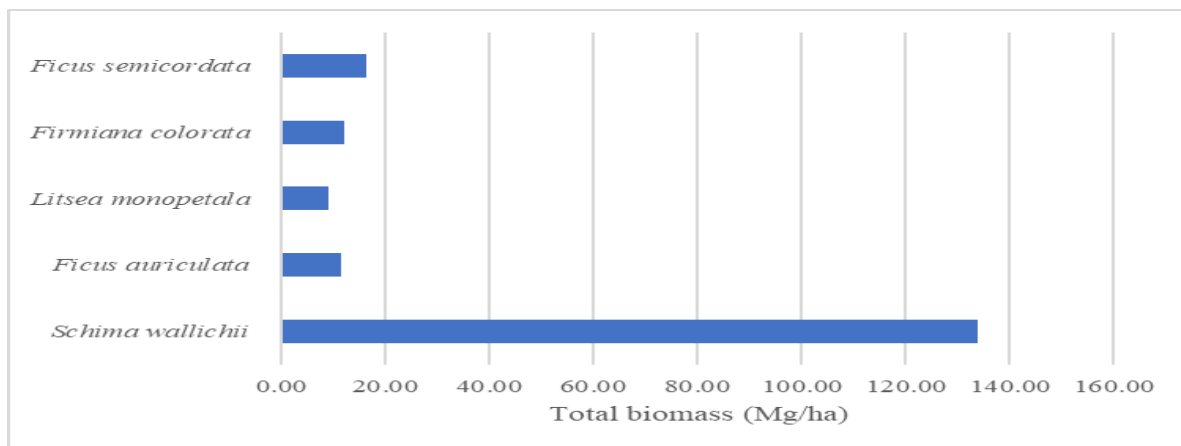
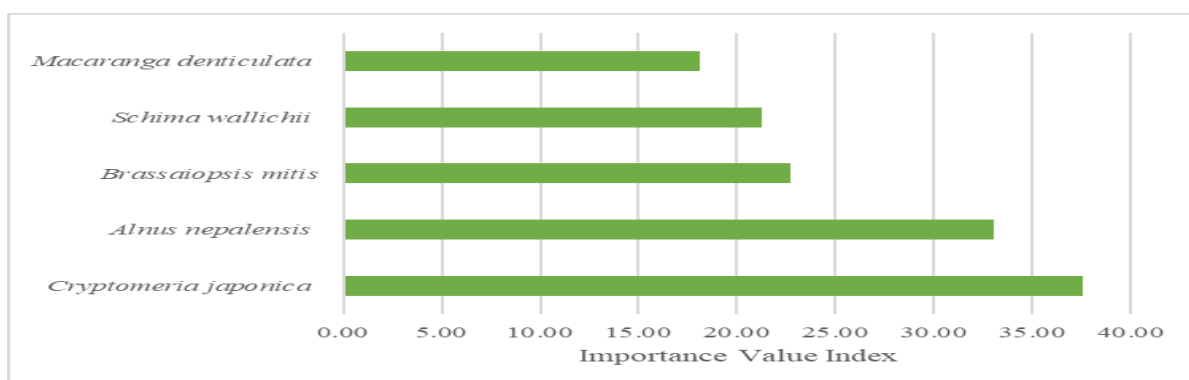


Fig 3. IVI value of 5 dominant species at altitude class 700-1200 m



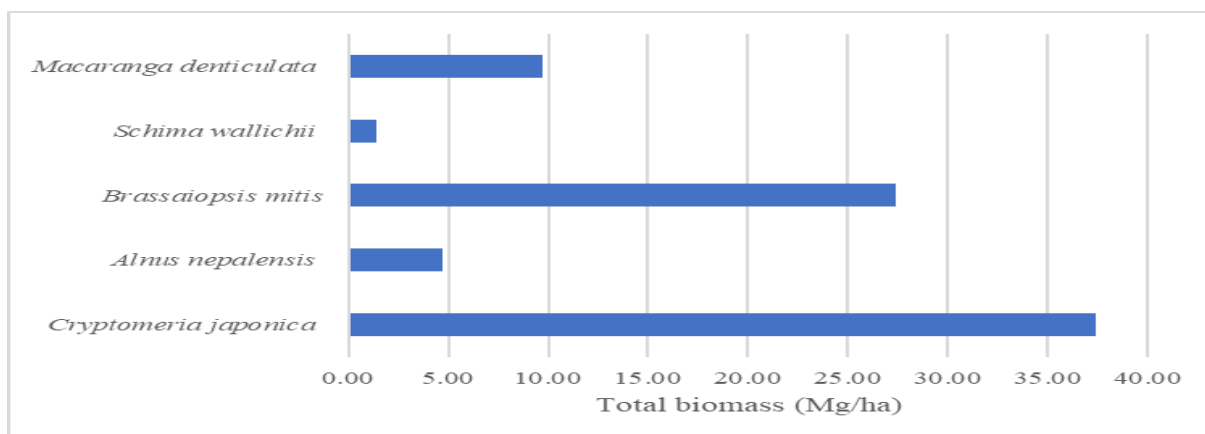
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939 Fig 4. Total biomass of 5 dominant species at altitude class 700-1200 m



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941 Fig 5. IVI value of 5 dominant species at altitude class 1200-1700 m



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943 Fig 6. Total biomass of 5 dominant species at altitude class 1200-1700 m

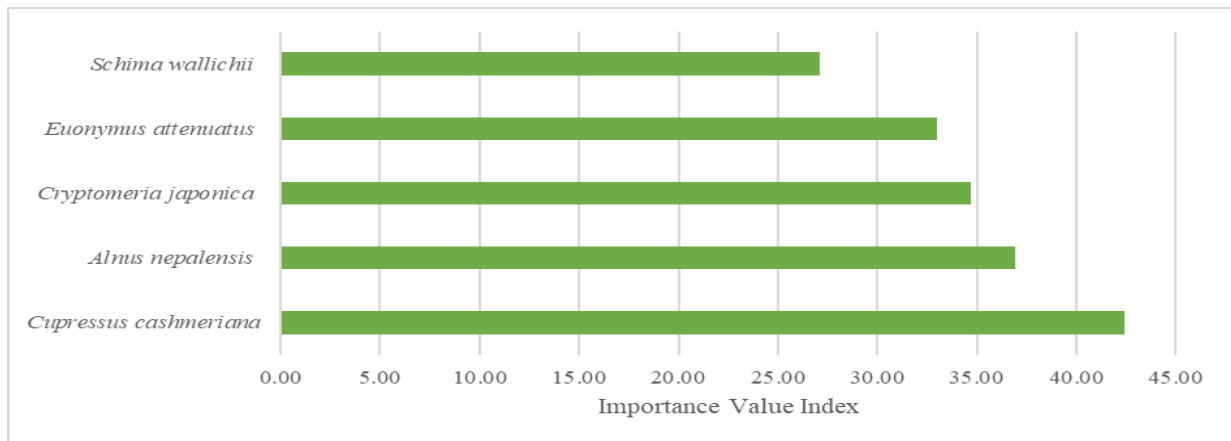


Fig 7. IVI value of 5 dominant species at altitude class >1700 m

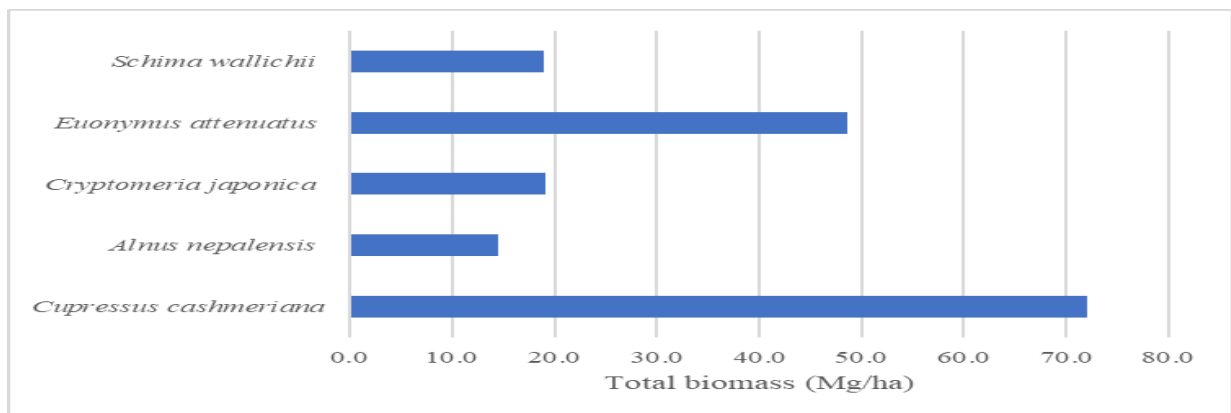


Fig 8. Total biomass of 5 dominant species at altitude class >1700 m

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