

Biomass Partitioning and Ecosystem Carbon Stocks of *Populus deltoides* and *Leucaena leucocephala* under Different High-Density Planting Spacings From North-Western India

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ABSTRACT

Agroforestry systems play a pivotal role in enhancing biomass production and carbon sequestration, thereby contributing to climate change mitigation and sustainable land management. The present study evaluated biomass accumulation and carbon stock of 8-year-old *Populus deltoides* and *Leucaena leucocephala* plantations established in 2013 under four high-density planting spacings (3.0×1.0 , 3.0×1.5 , 3.0×2.0 , and 3.0×2.5 m), using a split-plot design with three replications during 2021. Results revealed that aboveground biomass (AGB) was significantly higher in *Leucaena* (225.2 t ha^{-1}) than *Populus* (201.1 t ha^{-1}), whereas belowground biomass (BGB) was 73.3% greater in *Leucaena* (63.14 t ha^{-1}). Carbon concentration ranged from 43.28–49.29% among aboveground components and 44.48–46.25% among belowground components, with the highest values consistently observed in the bole and stump roots, respectively. Total aboveground carbon stock was significantly higher in *Leucaena* (109.7 t ha^{-1}) than *Populus* (97.91 t ha^{-1}), while belowground carbon stock was numerically greater in *Leucaena* (20.66 t ha^{-1}). Wider planting spacing ($3.0 \times 2.5 \text{ m}$) significantly enhanced biomass production and vegetation carbon stock, recording the highest total biomass (338.7 t ha^{-1}), vegetation carbon stock (165.9 t ha^{-1}), and total ecosystem carbon stock (217.6 t ha^{-1}). Conversely, soil carbon stock declined with increasing spacing and was highest under the closest spacing ($3.0 \times 1.0 \text{ m}$). Overall, under the 8-year-old plantation conditions evaluated in the present study, *Leucaena* demonstrated greater biomass production and carbon sequestration potential than *Populus*, while wider spacing promoted greater vegetation carbon accumulation. These findings highlight the importance of appropriate species selection and planting geometry for maximizing carbon sequestration and improving the climate change mitigation potential of high-density agroforestry systems.

1 | INTRODUCTION

Agroforestry has emerged as one of the most effective land-use systems for enhancing agricultural sustainability while mitigating climate change through biomass production and carbon sequestration (Jinger *et al.*, 2023; Dhobal *et al.*, 2024; Kaushal *et al.*, 2024). By integrating trees with crops, agroforestry systems capture atmospheric carbon in aboveground biomass, belowground biomass, and soil organic matter while simultaneously improving soil fertility, nutrient cycling, and ecosystem resilience (Jinger *et al.*, 2025; Gautam *et al.*, 2025). Depending on species, management, and environmental conditions, agroforestry systems can sequester $0.2\text{--}30.1 \text{ t C ha}^{-1} \text{ yr}^{-1}$, with the humid tropics exhibiting the highest carbon storage potential (Jose, 2009; Ghale *et al.*, 2022). Carbon stored in tree biomass generally ranges from 3 to 25 t C ha^{-1} , highlighting the

importance of selecting suitable tree species and management practices for maximizing ecosystem carbon storage (Jinger *et al.*, 2022; Kumara *et al.*, 2023).

Among the fast-growing tree species used in agroforestry, *Populus deltoides* and *Leucaena leucocephala* are widely cultivated because of their rapid growth, high biomass productivity, and adaptability to diverse agro-climatic conditions (Kumar *et al.*, 2021; Singh *et al.*, 2020). Poplar-based agroforestry systems are extensively adopted across northern India for timber production and carbon sequestration, whereas *Leucaena*, a nitrogen-fixing multipurpose tree, contributes substantially to biomass production, soil fertility improvement, and long-term carbon storage (Arora *et al.*, 2014; Chavan *et al.*, 2022). Besides increasing biomass, these species enhance soil organic carbon through litter deposition, root turnover, and

organic matter incorporation, thereby improving soil quality and sustaining long-term productivity (Kanuja and Bhatia, 2007; Chavan et al., 2022).

Planting spacing is a critical silvicultural practice that regulates tree growth, biomass partitioning, and carbon allocation by influencing competition for light, water, nutrients, and growing space. Wider spacing generally promotes greater individual tree growth, stem diameter, crown expansion, and biomass accumulation, whereas closer spacing may enhance soil carbon inputs through greater root density and litter deposition per unit area (Dowell et al., 2009; Prasad et al., 2011). Consequently, planting geometry affects not only biomass production but also the distribution of carbon among aboveground biomass, belowground biomass, and soil pools (Liu et al., 2018; Bahrian et al., 2024).

Although several studies have quantified biomass production and carbon sequestration in *Populus deltoides* (Arora et al., 2014; Chavan et al., 2023) or *Leucaena leucocephala* plantations (Prasad et al., 2011; Hanif et al., 2015) individually, comparative information on biomass partitioning and ecosystem carbon storage of these two fast-growing species under different high-density planting spacings remains limited. Furthermore, most previous studies have focused either on vegetation biomass or soil carbon alone, while integrated assessment of vegetation and soil carbon pools across the soil profile has received relatively little attention, despite its importance for accurately evaluating the carbon sequestration potential of agroforestry systems (Radrizzani et al., 2011; Conrad et al., 2017).

We hypothesized that (i) *Leucaena leucocephala*, owing to its rapid growth and nitrogen-fixing ability, would accumulate greater biomass and carbon stocks than *Populus deltoides*, and (ii) wider planting spacings would enhance aboveground and belowground biomass accumulation and vegetation carbon stock, whereas closer spacings would favour greater soil carbon storage. Therefore, the present study was undertaken to (i) quantify aboveground and belowground biomass production of *Populus deltoides* and *Leucaena leucocephala* under four planting spacings (3.0×1.0 , 3.0×1.5 , 3.0×2.0 , and 3.0×2.5 m), (ii) estimate carbon concentration and carbon stocks in different tree components and soil (0–30, 30–60, and 60–90 cm), and (iii) evaluate the influence of species and planting spacing on total ecosystem carbon storage in high-density agroforestry plantations.

2 | MATERIALS AND METHODS

2.1 | Experimental Site

The field experiment was conducted during 2021 at Agroforestry Research Centre, Haldi at Govind Ballabh Pant University of Agriculture and Technology, Pantnagar, Uttarakhand. The Centre is situated at 29° N Latitude, 79°30' E longitude and at an altitude of 243.84 m above the mean sea level, which lies in the foothills of the Shivalik

HIGHLIGHTS

- *Leucaena leucocephala* produced 11.98% higher aboveground biomass and 73.3% higher belowground biomass than *Populus deltoides*.
- Wider spacing (3.0×2.5 m) maximized biomass production, vegetation carbon stock (165.9 t ha^{-1}) and total ecosystem carbon stock (217.6 t ha^{-1}).
- Carbon concentration was highest in bole and stump roots, indicating greater carbon storage in woody components than in leaves and fine roots.
- Wider spacing enhanced vegetation carbon storage, whereas closer spacing increased soil carbon stock, demonstrating a trade-off between biomass and soil carbon sequestration.

range of the Central Himalayas and the North-Western Plain agro-climatic zone in the narrow strip called 'Tarai'. It falls in the sub-humid and sub-tropical climatic zone. The maximum daily temperature in summer may reach up to 42°C and minimum temperature in winter may fall up to 0.5°C. The monsoon sets in during the second or third week of June and continues until the end of September. Generally, the south-west monsoon sets in during the second or third week of June and continues until the end of September. The mean annual rainfall is about 1450 mm, of which 80-90 per cent is received during the wet season (July and September). The soils of "tarai" region are developed from calcareous, medium to moderately coarse textured materials under predominant influence of tall vegetation and moderate to well-drained conditions.

2.2 | Experimental Design

The experiment was conducted in a split-plot design (SPD) with three replications. The main-plot factor consisted of two tree species, viz., *Populus deltoides* and *Leucaena leucocephala*, while the sub-plot factor comprised four planting spacings: 3.0×1.0 m, 3.0×1.5 m, 3.0×2.0 m, and 3.0×2.5 m. The plantations were established in 2013, and the biomass and carbon stocks were assessed during 2021, when the trees were approximately 8-years-old. Thus, the experiment consisted of eight treatment combinations (2×4) arranged in three replications, resulting in a total of 24 experimental plots.

2.3 | Biomass Estimation

Tree biomass was estimated using the destructive sampling method. Three representative trees from each experimental plot were selected and destructively harvested during December. Consequently, a total of 72 trees ($2 \text{ tree species} \times 4 \text{ planting spacings} \times 3 \text{ replications} \times 3 \text{ trees per plot}$) were harvested for biomass estimation. Following harvesting, the aboveground biomass was separated into bole, branches, twigs, and leaves, and the fresh weight of each component was recorded immediately in the field. For belowground biomass estimation, root excavation was carried out in the western direction from the root collar. Roots were excavated along a transect extending 1.0 m horizontally from the tree centre to a depth of 1.5 m, covering an approximate soil volume of 1.5 m³. Coarse roots, comprising the stump root

and lateral roots, were excavated from this soil volume. Although a few coarse roots extended beyond the excavation boundary, their contribution was considered negligible. Lateral roots were defined as roots with a diameter ≥ 5 mm, whereas fine roots were defined as roots with a diameter ≤ 5 mm. Fine-root biomass was estimated by excavating three randomly distributed soil monoliths ($30 \times 30 \times 30$ cm) around each sampled tree. Fine roots below 30 cm depth were scarcely observed and were therefore not sampled. The excavated soil was dry-sieved manually, and roots were carefully hand-sorted to remove roots of other plant species. Representative samples (approximately 200 g) from each aboveground and belowground component were oven-dried at 65°C until constant weight. The dry biomass of each component was estimated using Equation 1 described by Husch *et al.*, (1972). Total biomass of *Populus* and *Leucaena* was calculated by summing the dry biomass of all aboveground and belowground components.

$$\text{Moisture content (\%)} = \frac{\text{Fresh weight of the sample (kg)} - \text{Oven dry weight of the sample (kg)}}{\text{Oven dry weight of the sample (kg)}} \times 100 \quad (\text{Eq. 1})$$

2.4 | Tree Carbon Stock

Carbon concentration in each biomass component was determined by the combustion (ash) method (Gallardo and Merino, 1993). Oven-dried samples were ground, and approximately 20 g of powdered material was combusted in a muffle furnace at $600 \pm 50^\circ\text{C}$ for 4–5 h. Carbon content was estimated from the ash-free dry matter, and carbon stock of individual tree components was calculated by multiplying dry biomass with the corresponding carbon concentration. Total vegetation carbon stock was obtained by summing carbon stored in all aboveground and belowground components.

2.5 | Soil Sampling and Carbon Stock Estimation

Soil samples were collected beneath the canopy of *Populus* and *Leucaena* plantations at four planting spacings from three soil depths: 0–30, 31–60, and 61–90 cm. The collected samples were air-dried, ground, passed through a 2-mm sieve, and analyzed for soil organic carbon (SOC) using the Walkley and Black (1934) wet oxidation method. Bulk density was determined at the corresponding soil depths using cylindrical metal core samplers (4.0 cm height $\times$ 5.0 cm internal diameter) following the method of Blake and Hartge (1986). The soil cores were oven-dried at $105 \pm 1^\circ\text{C}$ for 48 h to obtain constant weight. Bulk density (g cm^{-3}) was calculated as the oven-dry mass of soil divided by the internal volume of the core sampler. Soil organic carbon stock (Mg C ha^{-1}) for each depth was estimated by multiplying the SOC concentration (%), bulk density (Mg m^{-3}), and soil depth (cm), following equation 2 described by Paramesh *et al.* (2022) and Jinger *et al.* (2024). The total soil carbon stock was obtained by summing the carbon stock across the three soil depths (0–90 cm).

$$\text{SOC stock (Mg ha}^{-1}\text{)} = \frac{\text{SOC} \times \text{BD} \times \text{SD} \times 10000}{100} \quad (\text{Eq. 2})$$

Where,

BD is bulk density (Mg m^{-3}), SD is soil depth (m), and SOC is soil organic carbon (%).

2.6 | Statistical Analysis

The data were subjected to analysis of variance (ANOVA) appropriate for a split-plot design following the procedures described by Panse and Sukhatme (1967). The significance of treatment effects was tested using the F-test, and treatment means were compared using the Critical Difference (CD) at the 5% probability level.

3 | RESULTS AND DISCUSSION

3.1 | Above-Ground Biomass (AGB)

The AGB accumulation of *Populus* and *Leucaena* under different planting spacings is presented in Table 1. The contribution of individual aboveground components to total biomass followed the order: bole > branch > leaf > twig in both species. In *Populus*, the bole contributed 64.39% of the total AGB, followed by branches (13.45%), leaves (6.07%) and twigs (0.76%). Similarly, in *Leucaena*, the corresponding contributions were 58.71%, 20.69%, 3.27%, and 0.66%, respectively. Tree species significantly influenced branch and total AGB, whereas bole, leaf and twig biomass remained statistically comparable. *Leucaena* recorded significantly higher total AGB (225.2 t ha^{-1}) than *Populus* (201.1 t ha^{-1}), representing an 11.98% increase. Likewise, branch biomass was 75.1% higher in *Leucaena* (55.93 t ha^{-1}) than in *Populus* (31.95 t ha^{-1}). Planting spacing significantly affected the biomass production of all aboveground components except leaves. Total AGB increased progressively with wider spacing and was highest at $3.0 \times 2.5 \text{ m}$ (288.5 t ha^{-1}), which was statistically at par with $3.0 \times 2.0 \text{ m}$ (273.4 t ha^{-1}). The $3.0 \times 2.5 \text{ m}$ spacing increased total AGB by 131.7%, 73.5%, and 5.5% over $3.0 \times 1.0 \text{ m}$, $3.0 \times 1.5 \text{ m}$, and $3.0 \times 2.0 \text{ m}$, respectively. Similarly, bole biomass increased by 215.7%, 97.3%, and 4.5%, while branch biomass increased by 23.7%, 21.4%, and 7.8% compared with the respective spacings. Twig biomass was also significantly higher at $3.0 \times 2.0 \text{ m}$ (2.05 t ha^{-1}), followed closely by $3.0 \times 2.5 \text{ m}$ (1.89 t ha^{-1}). A significant tree species $\times$ spacing (A $\times$ B) interaction was observed for all AGB components, indicating that the response of biomass accumulation to planting spacing varied between *Populus* and *Leucaena*. In both species, the bole contributed the largest share of total biomass, followed by branches, leaves and twigs, which agrees with Liu *et al.* (2018), who reported that stem biomass constitutes more than 55.9% of the total AGB of *Leucaena*. Similar high biomass productivity of *L. leucocephala* has been reported under different agro-climatic conditions by Venilla *et al.* (2020). *Leucaena leucocephala* cv. Tarramba produced biomass of 26 kg tree^{-1} in two years under the cool winter conditions of Brisbane (Mullen and Gutteridge, 2002).

Wider planting spacings (3.0×2.5 and $3.0 \times 2.0 \text{ m}$) significantly enhanced biomass accumulation, probably due to reduced competition for light, moisture and nutrients,

resulting in greater stem growth and crown development. Similar responses have been reported in *Populus deltoides*, where improved plantation geometry and growing space increased tree height, diameter and biomass production (Chavan *et al.*, 2022). The high biomass production of poplar observed in the present study is also consistent with Chavan *et al.*, (2023), who reported biomass accumulation of approximately 180 t ha⁻¹ in poplar-based agroforestry

systems over a six-year rotation, while Chauhan *et al.*, (2010) recorded a 113.6% increase in total system biomass under poplar–wheat agroforestry compared with sole wheat. These findings indicate that species characteristics together with optimum planting spacing play a crucial role in maximizing biomass production in commercial agroforestry systems.

TABLE 1 Above and below ground biomass production (t ha⁻¹) as influenced by tree species and spacing under high density plantation.

Treatments	Above ground biomass production (t ha ⁻¹)					Below ground biomass production (t ha ⁻¹)			
	Bole	Branch	Leaf	Twig	Total	Stump root	Lateral root	Fine root	Total
<i>Tree species</i>									
<i>Populus</i>	152.9 (64.39)	31.95 (13.45)	14.43 (6.07)	1.81 (0.76)	201.1	22.43 (9.44)	12.36 (5.20)	1.63 (0.68)	36.43
<i>Leucaena</i>	158.6 (58.71)	55.93 (20.69)	8.84 (3.27)	1.81 (0.66)	225.2	27.93 (10.33)	16.53 (6.11)	0.49 (0.18)	63.14
C.D. (P=0.05)	NS	4.42	0.27	NS	14.8	NS	NS	S	9.57
<i>Tree spacing</i>									
3.0m x 1.0m	70.98 (43.51)	39.91 (24.46)	12.05 (7.38)	1.57 (4.97)	124.5	24.13 (14.76)	12.94 (7.93)	1.52 (0.93)	38.61
3.0m x 1.5m	113.6 (55.25)	40.68 (19.76)	10.23 (4.97)	1.74 (0.84)	166.3	23.21 (11.28)	15.21 (7.39)	1.09 (0.52)	39.52
3.0m x 2.0m	214.4 (69.62)	45.79 (14.86)	11.15 (3.62)	2.05 (0.66)	273.4	20.78 (6.74)	12.96 (4.20)	0.84 (0.27)	34.59
3.0m x 2.5m	224.1 (66.18)	49.37 (14.58)	13.10 (3.86)	1.89 (0.55)	288.5	32.60 (9.62)	16.75 (4.94)	0.81 (0.23)	50.17
C.D. (P=0.05)	8.85	1.95	NS	0.26	9.26	2.46	1.66	0.11	3.07
Interaction (AxB)	S	S	S	S	S	S	S	S	S

(Values in parentheses are the per cent contribution of different component of AGB and BGB for total biomass production) *S= Significant, NS= Non-Significant.

3.2| Below-Ground Biomass (BGB)

The BGB accumulation of *Populus* and *Leucaena* under different planting spacings is presented in Table 1. The contribution of BGB components followed the order: stump root > lateral root > fine root in both species. In *Populus*, stump root, lateral root, and fine root contributed 9.44%, 5.20%, and 0.68%, respectively, whereas the corresponding contributions in *Leucaena* were 10.33%, 6.11%, and 0.18% of the total biomass. Tree species significantly influenced total BGB and fine-root biomass, whereas stump-root and lateral-root biomass did not differ significantly between species. *Leucaena* produced significantly greater total BGB (63.14 t ha⁻¹) than *Populus* (36.43 t ha⁻¹), representing a 73.3% increase. Fine-root biomass, however, was significantly higher in *Populus* (1.63 t ha⁻¹) than in *Leucaena* (0.49 t ha⁻¹), indicating a 232.7% greater fine-root biomass in *Populus*. Planting spacing significantly affected all BGB components. The widest spacing (3.0 × 2.5 m) recorded the highest total BGB (50.17 t ha⁻¹), which was 29.9%, 27.0%, and 45.0% higher than 3.0 × 1.0 m, 3.0 × 1.5 m, and 3.0 × 2.0 m, respectively. Similarly, stump-root biomass at 3.0 × 2.5 m (32.60 t ha⁻¹) increased by 35.1%, 40.5%, and 56.9% over 3.0 × 1.0 m, 3.0 × 1.5m, and 3.0 ×

2.0 m, respectively. Lateral-root biomass was also highest at 3.0 × 2.5 m (16.75 t ha⁻¹), showing increases of 29.4%, 10.1%, and 29.2% over the respective spacings. In contrast, fine-root biomass declined with increasing spacing and was highest at 3.0 × 1.0 m (1.52 t ha⁻¹), decreasing by 28.3%, 44.7%, and 46.7% at 3.0 × 1.5 m, 3.0 × 2.0 m, and 3.0 × 2.5 m, respectively. A significant tree species × spacing (A × B) interaction was observed for all BGB components, indicating that the response of root biomass allocation to planting spacing differed between *Populus* and *Leucaena*. The significantly higher BGB of *Leucaena* than *Populus* indicates its superior root development and biomass allocation under the present agroforestry system. In both species, stump roots contributed the largest proportion of total BGB, followed by lateral and fine roots, which is consistent with the findings of Liu *et al.* (2018). The greater total BGB observed under the widest spacing (3.0 × 2.5 m) may be attributed to reduced competition for soil moisture, nutrients and rooting space, resulting in enhanced root growth. Similar responses of biomass production to stand density were reported by Prasad *et al.* (2011) in *Leucaena leucocephala* and Dowell *et al.* (2009) in *Populus deltoides*. Conversely, the higher fine-root biomass under closer spacing suggests an adaptive response to an increase in below-ground competition for resources. These findings are

further supported by Chavan *et al.* (2023), who highlighted the high biomass production and carbon sequestration potential of *Populus*- and *Leucaena*-based agroforestry systems, emphasizing the importance of optimum planting geometry for maximizing biomass accumulation.

3.3 | Carbon Content

The carbon concentration of different aboveground and belowground components of *Populus* and *Leucaena* is presented in Fig.1. Carbon concentration of the aboveground components ranged from 43.28 to 49.25% in *Populus* and 43.99 to 49.29% in *Leucaena*. In both species, the bole recorded the highest carbon concentration, followed by branch, twig, and leaves. In *Leucaena*, carbon concentration was highest in the bole (49.29%), followed by branch (48.31%), twig (44.57%), and leaves (43.99%). Likewise, in *Populus*, the bole contained the highest carbon concentration (49.25%), followed by branch (48.12%), twig (44.31%), and leaves (43.28%). Compared with leaves, carbon concentration in the bole was 14.5% higher in *Populus* and 12.0% higher in *Leucaena*, whereas branch carbon concentration exceeded that of leaves by 11.2% and 9.8%, respectively. Among the belowground components, carbon concentration ranged from 44.48 to 46.00% in *Populus* and 44.61 to 46.25% in *Leucaena*. In both species, the stump root recorded the highest carbon concentration, followed by the lateral root and fine root. In *Leucaena*, stump root carbon concentration (46.25%) was 1.8% and 3.7% higher than that of lateral roots (45.43%) and fine roots (44.61%), respectively. Similarly, in *Populus*, stump roots (46.00%) contained 2.2% and 3.4% higher carbon concentration than lateral roots (45.00%) and fine roots (44.48%), respectively. Overall, carbon concentration was consistently greatest in the bole among aboveground components and in the stump root among belowground components in both tree species, indicating greater carbon

storage potential in woody tissues than in leaves and fine roots. The carbon concentration of different components of *Populus* and *Leucaena* observed in the present study was within the normal range reported for fast-growing woody tree species. In both species, the bole exhibited the highest carbon concentration, followed by branches, twigs and leaves, indicating that woody tissues serve as the principal long-term carbon reservoir because of their higher lignin and cellulose content. Similar biomass carbon partitioning has been reported in *Leucaena*, where stem tissues contained the largest proportion of carbon owing to greater structural biomass accumulation (Liu *et al.*, 2018). Likewise, Arora *et al.*, (2014) reported that woody components of *Populus* plantations stored considerably more carbon than foliage, emphasizing the importance of stem biomass in carbon sequestration. Among the belowground components, stump roots contained higher carbon concentration than lateral and fine roots in both species. This may be attributed to greater lignification and longer tissue longevity of coarse roots, whereas fine roots are metabolically active and undergo rapid turnover. Similar observations have been reported for *Populus*, where stump and coarse roots constituted the major belowground carbon pool (Bahrian *et al.*, 2024). Furthermore, Villanueva-Lopez *et al.*, (2018) reported greater carbon storage in stems and coarse roots than in foliage and fine roots of *Leucaena*-based systems, confirming that woody tissues contribute disproportionately to long-term carbon storage. The close agreement between the present findings and previous studies suggests that carbon concentration is primarily governed by tissue composition and species characteristics rather than plantation management, while differences in biomass accumulation ultimately determine the overall carbon sequestration potential of *Populus*- and *Leucaena*-based agroforestry systems.

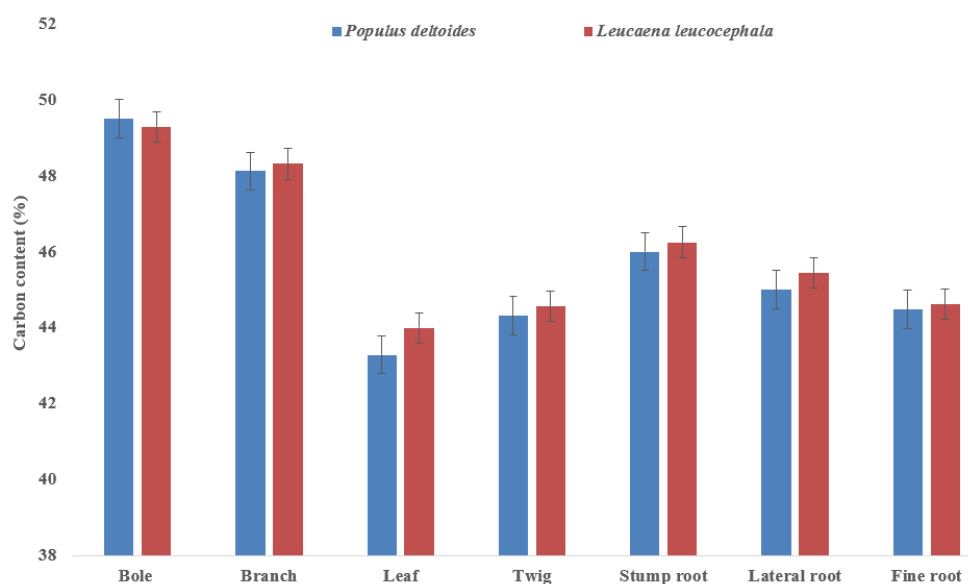


FIGURE 1 Mean carbon percentage (%) in different components of *Populus deltoides* and *Leucaena leucocephala*. Error bars indicate LSD ($p < 0.05$).

3.4 | Above-Ground Carbon Stock

The aboveground carbon stock of *Populus* and *Leucaena* under different planting spacings is presented in Table 2. The contribution of individual aboveground components to the total aboveground carbon stock followed the order: bole > branch > leaf > twig in both tree species. In *Populus*, the bole contributed 65.87% of the total aboveground carbon stock, followed by branches (13.49%), leaves (5.38%), and twigs (0.68%). Similarly, in *Leucaena*, the corresponding contributions were 60.11%, 20.64%, 2.79%, and 0.62%, respectively. Tree species significantly influenced branch, leaf, twig, and total aboveground carbon stock, whereas bole carbon stock remained statistically comparable. *Leucaena* recorded significantly higher total aboveground carbon stock (109.7 t ha⁻¹) than *Populus* (97.91 t ha⁻¹), representing a 12.0% increase. Likewise, branch carbon stock was 74.1% higher in *Leucaena* (26.92 t ha⁻¹) than in *Populus* (15.46 t ha⁻¹). In contrast, leaf carbon stock was 69.5% higher in *Populus* (6.17 t ha⁻¹) than in *Leucaena* (3.64 t ha⁻¹), while twig carbon stock differed only marginally between the two species. Planting spacing significantly influenced carbon storage in all aboveground components. Total aboveground carbon stock increased progressively with wider spacing and reached a maximum at 3.0 × 2.5 m (142.3 t ha⁻¹), which was 141.6%, 76.7%, and 6.5% higher than that recorded at 3.0 × 1.0 m, 3.0 × 1.5 m, and 3.0 × 2.0 m, respectively. Similarly, bole carbon stock increased by 226.0%, 102.5%, and 7.2% over the respective spacings. Branch carbon stock was highest at 3.0 × 2.5 m (24.10 t ha⁻¹), representing increases of 27.0%, 23.5%, and 8.7% over 3.0 × 1.0 m, 3.0 × 1.5 m, and 3.0 × 2.0 m, respectively. Leaf carbon stock remained statistically comparable among spacings, although numerically the highest value was recorded at 3.0 × 2.5 m (6.00 t ha⁻¹). Twig carbon stock was highest at 3.0 × 2.0 m (0.96 t ha⁻¹) and remained statistically comparable with 3.0 × 2.5 m (0.89 t ha⁻¹). A significant tree species × spacing (A × B) interaction was observed for all aboveground carbon stock components, indicating differential responses of *Populus* and *Leucaena* to planting spacing. The significantly higher aboveground carbon stock recorded in *Leucaena* than in *Populus* reflects its superior biomass production and carbon sequestration potential under agroforestry conditions. In both species, the bole constituted the largest carbon pool, followed by branches, leaves and twigs, indicating that woody tissues are the principal reservoirs of aboveground carbon. Similar observations were reported by Prasad et al. (2011), who found that the bole alone stored approximately 44 t C ha⁻¹, while total aboveground carbon stock reached about 50 t C ha⁻¹ and total ecosystem carbon stock was 62 t C ha⁻¹ in *Leucaena* plantations. Likewise, Hanif et al. (2015) reported that *Leucaena* accumulated about 135 Mg C ha⁻¹ in biomass and soil over seven years, whereas Sheikh et al. (2015) recorded approximately 62 Mg C ha⁻¹ carbon stock in four-year-old *Leucaena*-based farm forestry systems. Lopez-Santiago et al. (2019) further demonstrated that intensive *Leucaena* silvopastoral

systems stored 106.5 t CO₂-eq ha⁻¹ in above- and belowground biomass, highlighting the high carbon sequestration capacity of this species. The wider planting spacings (3.0 × 2.5 and 3.0 × 2.0 m) significantly enhanced aboveground carbon stock, primarily due to greater biomass accumulation under reduced competition for resources. Similar trends have been reported for *Populus*, where aboveground carbon stock increased with plantation age and stand growth. Reddy et al. (2025) reported that total plant carbon stock increased from 0.89 to 43.54 Mg ha⁻¹ between two and six years of age, while Kumar et al. (2023) observed a total carbon stock of 88.9 t C ha⁻¹, of which approximately 71.6 t C ha⁻¹ (80.5%) was stored in aboveground biomass. Similarly, Yasin et al. (2025) reported that six-year-old poplar clone G-48 accumulated 0.287 t C tree⁻¹, corresponding to about 129.8 Mg CO₂ ha⁻¹ mitigation potential. These findings corroborate the present study and demonstrate that both *Leucaena* and *Populus* are efficient carbon-sequestering tree species, with aboveground woody biomass serving as the major carbon sink under well-managed agroforestry systems.

3.5 | Below-Ground Carbon Stock

The belowground carbon stock of *Populus* and *Leucaena* under different planting spacings is presented in Table 2. The contribution of individual belowground components to total belowground carbon stock followed the order: stump root > lateral root > fine root in both species. In *Populus*, stump root, lateral root, and fine root contributed 9.04%, 4.88%, and 0.66%, respectively, whereas the corresponding contributions in *Leucaena* were 9.90%, 5.76%, and 0.16%, respectively. Tree species had no significant effect on total belowground carbon stock, stump-root carbon stock, or lateral-root carbon stock. However, fine-root carbon stock differed significantly between the species. Numerically, *Leucaena* recorded 23.4% higher total belowground carbon stock (20.66 t ha⁻¹) than *Populus* (16.74 t ha⁻¹). In contrast, fine-root carbon stock was significantly higher in *Populus* (0.76 t ha⁻¹) than in *Leucaena* (0.21 t ha⁻¹), representing a 261.9% increase. Planting spacing significantly influenced carbon stock in all belowground components. The widest spacing (3.0 × 2.5 m) recorded the highest total belowground carbon stock (23.69 t ha⁻¹), which was 37.7%, 31.9%, and 48.4% higher than 3.0 × 1.0 m, 3.0 × 1.5 m, and 3.0 × 2.0 m, respectively. Similarly, stump-root carbon stock at 3.0 × 2.5 m (15.52 t ha⁻¹) was 43.4%, 46.0%, and 61.3% higher than that recorded under 3.0 × 1.0 m, 3.0 × 1.5 m, and 3.0 × 2.0 m, respectively. Lateral-root carbon stock was also highest at 3.0 × 2.5 m (7.80 t ha⁻¹), showing increases of 36.6%, 14.2%, and 32.7% over the corresponding spacings. In contrast, fine-root carbon stock was highest at 3.0 × 1.0 m (0.66 t ha⁻¹) and declined with increasing spacing, decreasing by 25.8%, 31.8%, and 45.5% at 3.0 × 1.5 m, 3.0 × 2.0 m, and 3.0 × 2.5 m, respectively. A significant tree species × spacing (A × B) interaction was observed for all belowground carbon stock components, indicating that the response of belowground carbon allocation to planting spacing varied between *Populus* and *Leucaena*. The numerically higher belowground

nd carbon stock recorded in *Leucaena* than in *Populus* indicates the greater carbon allocation capacity of *Leucaena* to coarse root systems. In both species, stump roots accounted for the largest share of belowground carbon, followed by lateral and fine roots, highlighting the important role of coarse roots as long-term carbon reservoirs. Similar observations were reported by Bahrian *et al.* (2024), who demonstrated that stump and coarse roots contributed the major proportion of belowground carbon storage in *Populus* owing to their larger biomass and longer residence time. Likewise, Liu *et al.* (2018) reported that coarse roots represented the dominant belowground biomass component in *Leucaena*, whereas fine roots constituted only a small fraction because of their rapid turnover. The significantly higher belowground carbon stock under the widest spacing (3.0×2.5 m) suggests that

reduced competition for soil resources promoted greater coarse-root development and carbon accumulation. In contrast, the higher fine-root carbon stock observed under closer spacing may represent an adaptive response to greater competition for water and nutrients. Similar effects of stand density on root biomass and carbon allocation have been reported by Prasad *et al.* (2011) in *Leucaena*, where optimum stand density enhanced root biomass and carbon storage. Furthermore, Yasin *et al.* (2025) reported substantial belowground carbon accumulation in six-year-old *Populus* clone plantations, emphasizing the importance of root biomass in total ecosystem carbon sequestration. These findings indicate that both species possess considerable belowground carbon sequestration potential, and optimizing planting spacing can further enhance long-term carbon storage in agroforestry systems.

TABLE 2 Above and below ground carbon stock (t ha^{-1}) as influenced by tree species and spacing under high density plantation.

Treatments	Above ground carbon stock (t ha^{-1})					Below ground carbon stock (t ha^{-1})			
	Bole	Branch	Leaf	Twig	Total	Stump root	Lateral root	Fine root	Total
<i>Tree species</i>									
<i>Populus</i>	75.49 (65.87)	15.46 (13.49)	6.17 (5.38)	0.78 (0.68)	97.91	10.37 (4.88)	5.60 (4.88)	0.76 (0.66)	16.74
<i>Leucaena</i>	78.39 (60.11)	26.92 (20.64)	3.64 (2.79)	0.82 (0.62)	109.7	12.92 (9.90)	7.52 (5.76)	0.21 (0.16)	20.66
C.D. (P=0.05)	NS	2.20	0.18	0.058	7.10	NS	NS	0.04	NS
<i>Tree spacing</i>									
3.0m x 1.0m	34.75 (45.65)	18.97 (24.92)	4.60 (6.04)	0.54 (0.70)	58.90	10.82 (14.21)	5.71 (7.50)	0.66 (0.86)	17.20
3.0m x 1.5m	55.95 (56.81)	19.52 (19.82)	4.23 (4.29)	0.79 (0.80)	80.51	10.63 (10.79)	6.83 (6.93)	0.49 (0.49)	17.96
3.0m x 2.0m	105.7 (70.65)	22.17 (14.81)	4.78 (3.19)	0.96 (0.64)	133.6	9.62 (6.43)	5.88 (3.93)	0.45 (0.30)	15.96
3.0m x 2.5m	113.3 (68.29)	24.10 (14.52)	6.00 (3.61)	0.89 (0.53)	142.3	15.52 (9.35)	7.80 (4.70)	0.36 (0.21)	23.69
C.D. (P=0.05)	4.36	1.69	0.93	0.12	4.45	1.21	0.73	0.05	1.48
Interaction (AxB)	S	S	S	S	S	S	S	S	S

(Values in the parentheses are the per cent contribution of different component of above and below ground carbon stock for total carbon sequestration). *S= Significant, NS= Non-Significant

3.6 | Total Carbon Stock (t ha^{-1})

The vegetation, soil, and total carbon stock under *Populus* and *Leucaena* at different planting spacings are presented in Table 3. Tree species significantly influenced vegetation and total carbon stock, whereas soil carbon stock remained statistically unaffected. *Leucaena* recorded significantly higher vegetation carbon stock (130.4 t ha^{-1}) than *Populus* (114.6 t ha^{-1}), representing a 13.8% increase. Likewise, total carbon stock was significantly higher in *Leucaena* (194.3 t ha^{-1}) than in *Populus* (176.6 t ha^{-1}), reflecting a 10.0% increase. Although soil carbon stock was numerically higher under *Leucaena* (63.87 t ha^{-1}) than *Populus* (61.95 t ha^{-1}), the difference (3.1%) was statistically non-significant. Planting spacing had a significant effect on vegetation, soil, and total carbon stock. Vegetation carbon

stock increased progressively with wider spacing and reached a maximum at 3.0×2.5 m (165.9 t ha^{-1}), which was 118.0%, 68.5%, and 10.9% higher than that recorded at 3.0×1.0 m, 3.0×1.5 m, and 3.0×2.0 m, respectively. In contrast, soil carbon stock declined significantly with increasing spacing, with the highest value recorded at 3.0×1.0 m (80.02 t ha^{-1}) and the lowest at 3.0×2.5 m (51.69 t ha^{-1}), indicating a 35.4% reduction. Consequently, total carbon stock increased with wider spacing and was highest at 3.0×2.5 m (217.6 t ha^{-1}), representing increases of 39.4%, 34.7%, and 5.4% over 3.0×1.0 m, 3.0×1.5 m, and 3.0×2.0 m, respectively. A significant tree species $\times$ spacing (A $\times$ B) interaction was observed for vegetation and total carbon stock, whereas the interaction effect on soil carbon stock was non-significant. The significantly higher

vegetation and total carbon stock recorded in *Leucaena* than in *Populus* reflects the greater biomass production and carbon sequestration capacity of *Leucaena* under agroforestry conditions. Although soil carbon stock did not differ significantly between the two species, the slightly higher SOC under *Leucaena* may be attributed to greater litter input, biological nitrogen fixation, and enhanced belowground carbon inputs. Similar findings were reported by Radrizzani *et al.*, (2011), who observed that long-term *Leucaena*–grass pastures accumulated soil organic carbon and total nitrogen at rates of 267 and 16.7 kg ha⁻¹ yr⁻¹, respectively. Likewise, Conrad *et al.*, (2017) reported that nitrogen accretion from *Leucaena* (approximately 36 kg N ha⁻¹ yr⁻¹) promoted humus formation and increased surface soil organic carbon by 17–30% over time. The significant increase in vegetation and total carbon stock under wider spacing (3.0 × 2.5 m) was mainly associated with greater

tree biomass accumulation under reduced competition for light, water, and nutrients. In contrast, soil carbon stock was highest under the closest spacing, probably because of greater root density and continuous litter deposition per unit area. Similar improvements in soil carbon under *Populus*-based agroforestry have been reported by Chaudhari *et al.*, (2016), who observed 22.9% higher SOC under four-year-old poplar–wheat systems than sole wheat. Furthermore, Sirohi and Bangarwa (2017) demonstrated that *Populus* plantations improved SOC and nutrient distribution within the soil profile, thereby enhancing overall soil fertility. These findings indicate that while wider spacing maximizes vegetation carbon storage, closer tree spacing contributes more effectively to soil carbon accumulation, emphasizing the complementary role of planting geometry in optimizing ecosystem carbon sequestration.

TABLE 3 Total carbon stock (t ha⁻¹) as influenced by tree species and spacing under high density plantation.

Treatments	Total carbon stock (t ha ⁻¹)		
	Vegetation	Soil	Total
<i>Tree species</i>			
<i>Populus</i>	114.6	61.95	176.6
<i>Leucaena</i>	130.4	63.87	194.3
C.D. (P=0.05)	11.29	NS	13.25
<i>Tree spacing</i>			
3.0m x 1.0m	76.11	80.02	156.1
3.0m x 1.5m	98.47	63.04	161.5
3.0m x 2.0m	149.6	56.88	206.5
3.0m x 2.5m	165.9	51.69	217.6
C.D. (P=0.05)	4.60	4.83	6.62
Interaction (A x B)	S	NS	S

4 | CONCLUSIONS

The study demonstrated that both tree species and planting spacing significantly influenced biomass production and carbon sequestration under high-density agroforestry plantations. *Leucaena leucocephala* consistently outperformed *Populus deltoides* in terms of aboveground biomass, belowground biomass, vegetation carbon stock, and total ecosystem carbon stock, highlighting its superior carbon sequestration potential. In both species, the bole constituted the largest proportion of aboveground biomass and carbon, whereas stump roots contributed the greatest share of belowground biomass and carbon. Carbon concentration was consistently higher in woody tissues than in leaves and fine roots, indicating greater long-term carbon storage in structural components. Planting spacing markedly affected biomass partitioning and carbon accumulation. The widest spacing (3.0 × 2.5 m) produced the highest vegetation biomass and total carbon stock, whereas the closest spacing (3.0 × 1.0 m) stored more carbon in the soil. These results indicate a trade-off between aboveground biomass production and soil carbon storage with changing tree density. Overall, *Leucaena leucocephala* planted at wider spacing emerged as the most promising option for maximizing biomass production and carbon

sequestration in high-density agroforestry plantations. The findings provide valuable information for designing agroforestry systems aimed at enhancing carbon storage, improving ecosystem services, and supporting climate-resilient land management in the Tarai region and similar agroecological conditions.

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CONFLICT OF INTEREST

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

DATA AVAILABILITY STATEMENT

Data will be made available on request.

AUTHOR'S CONTRIBUTION

Seema Maikhuri, Sumit Chaturvedi, Sumit Gaur: Conceptualization, methodology, writing original manuscript; Seema Maikhuri, Sumit Chaturvedi, Sambita Bhattacharyya, Vipin Chandra Dhyani, Gurvinder: Lab

analysis, data collection; Statistical analysis; Seema Maikhuri, Sumit Chaturvedi, Sumit Gaur, Sambita Bhattacharyya, Vipin Chandra Dhyani, Gurvinder Singh, Rajesh Kaushal, Sujeet Kumar Lavania: Monitoring and supervision; Rajesh Kaushal, Sujeet Kumar Lavania, Dinesh Jinger: Interpretations, writing and reviewing.

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