

# Dynamics of ecosystem carbon in different forest types in the central Himalaya: Role of nitrogen-fixing Nepalese alder (*Alnus nepalensis* D. Don.)

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## Abstract

Nitrogen-fixing Nepalese alder (*Alnus nepalensis* D. Don.) is a fast-growing early successional species which often forms pure stands in areas affected by landslides and sometimes it occurs mixed with other species in the central Himalayas. In this study, we assessed the distribution of ecosystem carbon storage in plants and soil in a chronosequence of *A. nepalensis* forest stands in central Himalaya. We examined six forest stands: (1) *A. nepalensis*-early regeneration (AER) forest, (2) *A. nepalensis*-late regeneration (ALR) forest, (3) *A. nepalensis*- mature oak mixed (AMOM) forest, (4) *A. nepalensis*- mix with rhododendron (AMR) forest, (5) *A. nepalensis* –mix with old oak (AMOO) forest and (6) *A. nepalensis*-planted by the forest department in the degraded forest (APDF). The ecosystem C stock increased with an increase in stand total basal area (TBA). C storage in *A. nepalensis* tree biomass in different stand AER, APDF, ALR, AMOM, and AMR, AMOO, was 8.97, 51.41, 16.07, 53.74, 144.77, and 101.14 Mg ha<sup>-1</sup>, respectively. Soil organic C (SOC) in different soil depths in successional stages AER (0-10 cm), APDF (0-30 cm), ALR (0-100 cm), AMOM (0-100 cm), AMR (0-100 cm), and AMOO (0-100 cm) was 3.31, 31.21, 75.47, 157.04, 159.43 and 210.13 Mg ha<sup>-1</sup>, respectively, with decrease in SOC concentration with increasing soil depth. The ecosystem C storage averaged 15.85, 183, 216.26, 390.32, 403.66, and 500.08 Mg ha<sup>-1</sup> in AER, APDF, ALR, AMOM, AMR, and AMOO sites, respectively. Overall, in *A. nepalensis* forest development markedly ameliorated both vegetation and soil succession in central Himalaya.

## INTRODUCTION

Forest ecosystems play an important role in global biogeochemical cycles and mitigate the negative impact of climate change (Hui et al., 2017). Furthermore, forests act as a key carbon pool (C) and store more carbon per unit area than any terrestrial ecosystem (Dixon et al., 1994; Pugh et al., 2019). Figures suggest that half of the carbon in the terrestrial ecosystem is stored in forests (Pan et al., 2011; Popkin, 2019). Forest stand growth can change the forest structure, biomass, and soil nutrient content (Shanin et al., 2014; Goebes et al., 2019). The distribution of carbon stocks in different elements of ecosystems is one of the important features of succession (Robinson et al., 2015; Badalamenti et al., 2019). Natural vegetation in freshly degraded land/slip and the naturally regenerated forest is an effective way to deter soil degradation, tree establishment, improve the ecological environment, and drives succession (Walker, & del Moral2009; Berrahmouni et al., 2015; Pandit et al., 2018). Early succession species have strongly favored by vegetation restoration efforts due to their capability of tolerating extremely harsh environmental conditions (Walker, & del Moral2009). After forest degradation, early successional species invade the habitat and radically alter vegetation and soil physicochemical properties (Lebrija-Trejos et al., 2010). Early-successional forest ecosystems that grow after forest degradation or stand- replacement are very important, particularly in terms

of carbon storage by pioneer trees after landscape recovery (Swanson et al., 2010; Preem et al., 2012; Lorenc-Plucińska et al., 2013; Becker et al., 2015). The ability of *Alnus* tree to fixing nitrogen from the air through symbiotic with nitrogen-fixing bacteria, collectively called *Frankia*, generates to the concept that this tree could be effective agents to tolerating extremely harsh environmental conditions and accelerate the natural succession. *Alnus* is found mostly in degraded habitats with nutrient impoverished conditions in the soil (Sharma et al., 1998; Resh et al., 2002; Binkley., 2003; Perakis and Pett-Ridge 2019). Additionally, alder also produces cluster roots which secrete carboxylates and mobilizes available phosphorus and other mineral elements in the soil (Lambers et al., 2019).

Nitrogen-fixing Nepalese alder (*Alnus nepalensis* D. Don.) being an early successional species arises as a pioneer in post-disruption (landslides/ landslips) in most Himalayan forests below 2500 m (Sharma et al., 1998; Rana et al., 2018; Joshi and Garkoti 2020). In the central Himalaya, *A. nepalensis* often forms pure patches and sometimes grows in association with white oak (*Quercus leucotrichophora* A. Camus) as well as other broad-leaved species throughout different stages of forest succession after disturbance (Singh, 2014; Frouz et al., 2015; Joshi and Garkoti 2020). Owing to the association of *Frankia* in roots, the *Alnus* species fix nitrogen and have a stronger impact on soil physicochemical characteristics (Binkley et al 1992; Myrold et al., 1994; Resh et al., 2002; Binkley., 2003; Khan et al., 2007; Bissonnette et al., 2014; Perakis and Pett-Ridge 2019; Krishna et al. 2019). Therefore, it is widely used in agroforestry, forest management, and restoration systems and also has long been traditionally used as an intercropping tree species (Semwal et al., 2013; Sakalli et al., 2013; Sakalli et al., 2017; Rana et al., 2018). Despite a large series of studies addressing the advantages of *Alnus* species, knowledge on biomass C storage variations in alder species or alder mixed forest ecosystems in the central Himalayas is still limited. The proposed work envisages the role of *A. nepalensis* in the recovery of degraded ecosystems in central Himalaya. In the present study, a comprehensive analysis of the carbon storage in soil and vegetation along a chronosequence in *A. nepalensis* forest stands in central Himalaya is presented. The study was conducted to determine: (1) assess the above-and below-ground carbon storage of different *A. nepalensis* forest stands in central Himalaya (2) estimate carbon stock dynamics in the whole ecosystem (plant, litter, and soil) in of different *A. nepalensis* forest stands and (3) evaluate the role of *A. nepalensis* in influencing total ecosystem C pools as well as provide helpful suggestions on how to manage forests to mitigate impacts of climate change.

## 2. MATERIALS AND METHODS

### 2.1. Study area

The study was carried out at the Kedarnath valley of Rudraprayag district (30° 31. 44.7" N, 79° 6' 21.1" E and altitude 1604 masl) of Central Himalaya (Figure 1). The study area has a generally cold temperate type and a strongly seasonal climate. The mean annual precipitation ranged from 1971 mm, and most of this fall between June through September with moderate to heavy snowfall during December-February. The study area falls in the external zone of the Kedarnath wildlife sanctuary. The study area has steep hills along with undulating topography and influenced by landslide/slips during the rainy season. The soil is brown-black in color and podzolic, which is generally gravelly and large boulders are present in the area (Table.1). The study sites lie within the central axis of central Himalaya, which consists of belts of metamorphic rocks along with granites, gneiss, and schist referred to as the critical crystalline (Raina and Gupta 2009). The primary tree species in the study area are *Alnus nepalensis* D. Don, *Quercus leucotrichophora* A. Camus, *Rhododendron arboreum* Smith, *Myrica esculenta* Buch.-Ham, *Litsea umbrosa* Nees, *Lyonia ovalifolia* (Wall.) Drude, *Symplocos paniculata* Miq, *Pyrus pashia* L (Table.1).

### 2.2. Experimental design and sampling

In October 2018, six forest stands (1) *A. nepalensis*- early regeneration (AER) forest, (2) *A. nepalensis*-late regeneration (ALR) forest, (3) *A. nepalensis* -planted by the forest department in the degraded forest (APDF) (4) *A. nepalensis* - mature oak mixed (AMOM) forest, (5) *A. nepalensis* - mix with rhododendron (AMR) forest, and (6) *A. nepalensis* -mix with old oak (AMOO) forest in a chronosequence of *A. nepalensis* forest stands were randomly selected from the entire area of the Kedarnath valley. Proportion to the total

basal area of *A. nepalensis* of the stand was the primary criteria for stands selection. In each sampling stand, three 0.1 ha plots were established using a stratified random sampling design. Each plot was subdivided into ten (10 m x 10 m sized) quadrats for biomass and carbon inventory. Species density and tree diameter at breast height (DBH) were recorded in each plot. Tree biomass, including the biomass of bole, branch, twig, foliage, catkin, stump root, lateral root, and fine roots was estimated in plots by already developed allometric equations (Rawat, & Singh, 1988; Adhikari, 1994; Sharma & Ambhasht 1990; Sharma, 2011). Three 5 m x 5 m quadrats were layed to study Shrubs and three 1 m x 1 m sized quadrats were used for herbs and litter. Shrubs were harvested and separated into branches, roots, and leaves; herbs were harvested and separated into above-and belowground components from each subplot. Components of shrubs, herbs, and litter were brought to the laboratory and oven-dried at 64 °C for 48 h and weighed to estimate the ratio of fresh weight to dry biomass ( $\text{Mg ha}^{-1}$ ). The total vegetation C stock was computed by assuming that carbon content is 47.4% of the total biomass (Martin and Thomas, 2011). Soil samples were randomly collected with soil corers (diameter, 5 cm) at up to maximum soil depths (0-10 cm in AER, 0-30 cm in APDF, 0-100 cm in ALR, AMOM, AMR and AMOO plots) and divided in 0-10, 10-20, 20-30, 30-50, 50-80 and 80-100 cm. The soil samples were placed in plastic bags and brought to the laboratory. The soil sample was air-dried, and visible plant debris and stones were removed and ground to pass through a 2-mm sieve to perform organic carbon analysis. Soil organic carbon (SOC) was calculated using  $\text{K}_2\text{Cr}_2\text{O}_7\text{-H}_2\text{SO}_4$  oxidation (Nelson & Sommers, 1996). Bulk density was estimated for each soil depth by weighting the whole sample and oven-dried at 105 °C for 48 h.

Soil carbon stock (SOCS) was calculated according to the following equations:

$$\text{SOC stock } (\text{Mg ha}^{-1}) = \text{SOC} \times \text{BD} \times D \quad (1)$$

Where SOC (%) is soil organic carbon, D is the sampling depth (cm), BD is the bulk density ( $\text{g cm}^{-3}$ ) and SCFs is the mass of proportion of coarse fragments content (% of volume) using an average rock density of  $2.65 \text{ g cm}^{-3}$ .

Topographic feature (slope aspect, elevation, slope position, slope angle) of each plot was measured and slope correction was applied for final biomass estimation. Stands located on slope >10%, slope correction was applied using the following equation:

$$L = L_s \times \cos S \quad (2)$$

Where L is the true horizontal distance of forest stands  $L_s$  is the standard distance measured in the field along the slope and S is the slope in degrees. The area of the sampled stands was then calculated as: Area = stands width  $\times$  calculated true stands length (L).

## 2.3. Statistical analysis

All the results are reported as mean (three measurements)  $\pm$  standard error (SE) and statistical analysis performed using the R studio and excel 2013 software. C storage of vegetative components and soils, among the six forest stands, were compared using one-way ANOVA. Significance levels were set at  $\alpha=0.05$  for all statistical analyses.

## RESULT

### 3.1. Spatial variation of forest stand characteristics

The total basal area (TBA), species density, and DBH were significantly different across the stands. In contrast, TBA of different stands was lowest in AER ( $5.2 \pm 0.1 \text{ m}^2 \text{ ha}^{-1}$ ) and highest in AMOO ( $88.39 \pm 4.88 \text{ m}^2 \text{ ha}^{-1}$ ) ( $P < 0.05$ ). In AMR and AMOO forest stands, the stem sizes distributed among upper diameter classes

of 20-50 cm and >50 and resulted in the higher TBA ( $87.90 \text{ m}^2 \text{ ha}^{-1}$  and  $88.39 \text{ m}^2 \text{ ha}^{-1}$ , respectively). A large heterogeneity of tree diameter was observed among the stands (Figure 2). Results confirmed the dominance of *A. nepalensis* that occurred almost exclusively in the DBH category [?]30 cm except in AER stand. All the studied stands were predominantly composed of three species *A. nepalensis*, *Q. leucotrichophora*, and *R. arboreum*. The most abundant species by the number of stems was *Q. leucotrichophora* and *R. arboreum* and comprised 40-80 % of all stems. The proportion of individuals in the small-diameter class (<30 cm DBH) was higher in AER, ALR, APDF, and AMOM stand as compared to AMR and AMOO stands, where the proportion of individuals in the largest diameter class (>30 cm DBH) was higher (Table 2). Stand density was higher for young AER and ALR stands which declined in mature and older (AMR and AMOO) stands. In contrast, the stem diameter of AMOM stand was mostly confined within the diameter ranges of 10- 30 cm (Figure 2).

Species in the medium and large DBH classes were the primary contributors to tree biomass carbon storage in AMOM, AMR, and AMOO stands whereas the small DBH class was the primary contributor to biomass carbon in AER, ALR and APDF stand (Table 2). Although a majority of biomass carbon storage was contributed by trees of medium and large DBH classes in AMOM, AMR, and AMOO stand, the presence of a higher number of small trees in AER and ALR stand indicate for higher capacity of forest regeneration and thus a great potential for carbon accumulation. The higher contribution of large *A. nepalensis* trees to the biomass carbon found in the present study. In AER, APDF and ALR stand there were higher small trees than AMOM, AMR, and AMOO stand leading to a proportionally greater contribution of larger *A. nepalensis* trees to ecosystem carbon pools.

### Dynamics of biomass carbon storage

Tree biomass carbon and ecosystem biomass carbon of *A. nepalensis* stands increased with stand basal area ( $p < 0.01$ ) (Table 3). Based on the already developed allometric equations, the biomass of tree components was estimated (Table 4). Different tree components bole, branch, twig, foliage, stump root, lateral root, and fine root showed the same trend with an increase in stand basal area and followed the order of increase AER>ALR>APDF> AMOM>AMR>AMOO (Figures 3a, b). Total biomass carbon (tree, shrubs, herbs, and litter biomass carbon) and ecosystem biomass carbon increased from  $12.54 \text{ Mg ha}^{-1}$  to  $15.85 \text{ Mg ha}^{-1}$ , respectively, in AER stands to  $289.85 \text{ Mg ha}^{-1}$  to  $500 \text{ Mg ha}^{-1}$ , respectively in AMOO stand. Across the stands examined, tree bole biomass carbon contributed more to total tree biomass carbon than any other tree components. The understory vegetation (herbs and shrubs) and litter carbon in AER, ALR, APDF, AMOM, AMR, and AMOO stands were 3.06, 4.49, 7.14, 6.02, 7.92 and  $6.36 \text{ Mg ha}^{-1}$ , respectively. The litter biomass carbon increased with an increase in stand basal area, peaking in the AMR stands at  $3.56 \text{ Mg ha}^{-1}$  (Figure 3a).

The contribution of *A. nepalensis* to total biomass carbon was higher in APDF and AMR stands whereas the contribution of *Q. leucotrichophora* to biomass C was higher in the rest of the stands. The proportion of the bole and branch biomass carbon was the highest in AMR or AMOO and lowest in APDL. Fine root biomass carbon was lowest in APDL and highest in the AMOM while catkin biomass carbon contribution was lowest in all stands (Table 4). The litter and total ecosystem biomass showed a positive correlation with the forest basal area. The understory vegetation (herbs and shrub) did not show any trend with forest basal area. Moreover, the total biomass carbon of trees was always higher than that in the understory vegetation and litter biomass carbon. The highest above ground biomass carbon was found under AMOO ( $231.5 \text{ Mg ha}^{-1}$ ) followed by AMOM ( $183.58 \text{ Mg ha}^{-1}$ ) and belowground biomass carbon under AMOO ( $55.2 \text{ Mg ha}^{-1}$ ) followed by AMOM stand ( $54.9 \text{ Mg ha}^{-1}$ ).

### Dynamics of soil organic carbon storage

In all the stands, the SOC (%) decreased significantly with an increase in soil depths, while soil bulk density showed the reverse trends (Fig.4). The SOC % at 0-10 cm was higher than that of other soil depths. Average total soil C stock in AER (0-10 cm), APDF (0-30 cm), ALR (0-100 cm), AMOM (0-100 cm), AMR (0-100 cm), and AMOO (0-100 cm) was 3.31, 31.21, 75.47, 157.04, 159.43 and  $210.13 \text{ Mg ha}^{-1}$ , respectively (Figure

4).

### Change in ecosystem carbon stock

The ecosystem carbon stock increased significantly with stand basal area from 15.85 Mg ha<sup>-1</sup> in the AER with TBA 5.2 m<sup>2</sup>ha<sup>-1</sup> to 500.09 Mg ha<sup>-1</sup> in the AMOO stand with TBA 88.3 m<sup>2</sup> ha<sup>-1</sup>. Tree biomass carbon stock, soil organic carbon stock, and ecosystem carbon stock showed a positive correlation with stand basal area ( $P < 0.01$ ) and *A. nepalensis* basal area ( $P < 0.01$ ) (Figure 5a). The biomass carbon along with soil organic carbon increased with an increasing basal area of the stand (Figure 5b). The relationship between stand basal area and ecosystem carbon storage was  $y = 4.96254x + 27.534$  ( $R^2 = 0.8959$ ) was developed. In the case of *A. nepalensis* basal area to ecosystem carbon storage was,  $y = 6.8711x + 91937$  ( $R^2 = 0.9032$ ), and slope indicate that for the increase in *A. nepalensis* basal area, ecosystem carbon storage increased. Similarly, stand basal area and *A. nepalensis* basal area showed a positive increment with tree biomass carbon storage and SOC storage (Figure 5). Across the study sites, the percentage of biomass carbon contributed by *A. nepalensis* to the total ecosystem carbon storage ranged from 7.07 % (in ALR stand) - 63.20 % in AER stand). Similarly, *Q. leucotrichophora* contributed 5.07- 26.95 %, *R. arboreum* 1.43-19.36 % and associated species 8.31-21.58 % to the total ecosystem C stock. The shrubs, herbs, and litter contributed a small portion to the total ecosystem C stock of shrubs, and herbs C stock did not vary with stand basal area whereas litter biomass carbon showed a linear increase with a basal area of stands (Figure 6a). The vegetation biomass carbon storage contributed 77.08 %, 82.20 %, 63.92 %, 58.95 %, 59.62 % and 57.31 % of the total ecosystem carbon stock to the AER, APDF, ALR, AMOM, AMR, and AMOO stands respectively and the contribution of soil organic carbon stock to ecosystem carbon storage increased with the total basal area and ranged 20.89 % for APDF to 42.02 % in AMOO stand, indicating an increase in the soil carbon stock with the increase in the basal area of forest and *A. nepalensis* basal area (Figure 6b). Tree and soil were the two largest contributors to the total ecosystem carbon in all the stands. Ecosystem carbon storage in AER and APDF was low compared to other stands and ecosystem carbon stock significantly increased with the increase in *A. nepalensis* basal area.

## DISCUSSION

### Stand characteristics and biomass carbon storage in the tree, understory and forest floor

The basal area recorded in study stands (5.2-88.39 m<sup>2</sup>ha<sup>-1</sup>) is similar or higher than that reported for other oak forests in central Himalaya (Rawat & Singh 1988; Verma and Garkoti 2019). It has been observed that the stand basal area has a significant impact on stand productivity and carbon storage. The results of this study clearly illustrated that the stand-basal area reveals a strong correlation with the stand carbon stock in *A. nepalensis* stands (Figure 5). Our results indicate that there were significant differences in the biomass carbon storage in tree layer components across the chronosequence which indicate that; during the forest stand development, trees absorb atmospheric CO<sub>2</sub> and store it in plant structural parts. Biomass carbon availability mainly depends on dominant tree species. The structure of plant species can affect the storage of biomass carbon in the forest ecosystem (Conti and Diaz 2013). Our study underlines the importance of fast-growing and nitrogen-fixing *A. nepalensis* tree as a driver of the C cycle in the studies central Himalaya forest and includes detailed evidence of medium-and larger diameter classes trees were the primary contributor to above and below-ground biomass carbon storage. The carbon storage of the 3 most dominant species (*A. nepalensis*, *Q. leucotrichophora*, and *R. arboreum*) contribute for 63.50 % to 83.17% of the total biomass carbon storage and the *A. nepalensis* alone, accounted 10.77 to 69.19 % to the total biomass carbon storage, demonstrating that the majority of the biomass carbon storage is influenced by the *A. nepalensis* in the studies central Himalaya. The larger trees (DBH > 30 cm) contribute more to the basal area and thus in biomass carbon storage rather than total stem density. The larger size and fast-growing nature of *A. nepalensis* helped a greater amount of carbon storage. Variation in the density of large-diameter *A. nepalensis* to be impotent for storing more carbon stocks in forests. Results from our study agree with those of previous studies suggesting that forest stand structure playing an impotent role affecting biomass carbon storage in the forest. Large trees are the main contributors to forest biomass and ecosystem functions (Lutz et al., 2012; Lutz et al., 2018).

Other studies have also demonstrated that in matured forests large trees contain a large proportion of above-ground biomass (Slik et al., 2013; Bradford and Murphy 2019). These results cause us to predict that *A. nepalensis* will function as strong organizers of forest stand structure. This is an important discovery, as this tree is a more important and main source of biomass carbon storage. As a result, biomass carbon storage expanded as the basal area expanded and the related biomass carbon storage increased. The stand-basal area has a positive impact on the carbon storage of the forest ecosystem. Further, we anticipate that the being pioneer tree species itself, *A. nepalensis* showed good regeneration in AER stands and showing facilitative effects of this species on their neighbor's late succession *Q. leucotrichophora* and *R. arboreum* species. Plant community structure played a significant role in the storage of biomass carbon. In this study, ecosystem carbon storage was influenced by

*A. nepalensis*.

Our estimates of stands biomass across the basal area gradient demonstrated that tree biomass increased with the basal area of stands. Meanwhile, our results revealed present biomass estimation (10.02-284.48 Mg ha<sup>-1</sup>) was on the range recorded by earlier work on the oak forest in central Himalaya. Tree biomass carbon storage of AMOM, AMR and AMOO stands (228.05, 237.5, 284.48 Mg ha<sup>-1</sup> respectively) were same as other central Himalayan oak forests, while tree biomass carbon storage of AER, APDF and ALR stands (10.02, 148.28, 134.96 Mg ha<sup>-1</sup> respectively) were much lower than the central Himalaya oak forest (Rawat & Singh 1988; Adhikari et al., 1995; Sharma et al., 2011). We found AMOM, AMR, and AMOO stand had, on average, higher biomass carbon than other stands, supporting our hypothesis that suggests *A. nepalensis* abundance is a key driver of biomass storage in the central Himalaya forest. However, despite that studies stands may follow multiple succession pathways, our study suggests that succession trajectory may have a strong effect on total biomass carbon storage (Gough et al., 2016). In the present study, the aboveground biomass storage/belowground biomass storage ratio increased with a stand basal area this indicates that below ground carbon storage positively correlated to the basal area. Understory vegetation (shrubs, herbs) biomass carbon not increased with a basal area of stands but litter biomass carbon increased with stand basal area. The understory vegetation (shrubs and herbs) biomass carbon ranged from 2.11-3.26 Mg ha<sup>-1</sup>, which is the same as reported for the central Himalayan oak forest (Rawat & Singh 1988; Adhikari et al., 1995). Litter biomass carbon ranged from 0.32-3.56 Mg ha<sup>-1</sup> were close to reported values for oak forest litter biomass in the central Himalayan forest (Rawat & Singh 1988; Adhikari et al., 1995; Joshi & Garkoti 2020).

## 4.2. Soil organic content and storage

Throughout the stand growth, we significant increases in soil depth, which, in part, increased soil carbon pool. Our results showed the progressive increase in soil organic carbon storage proceeding from, younger to mature stages. In our results, the soil carbon contents and storage increased as the stand and *A. nepalensis* basal area increased, which indicates that *A. nepalensis* has a positive impact on the soil carbon content and storage. With forest development along the chronosequence, soil carbon and storage increased, which might be due to the nitrogen-rich litter of *A. nepalensis*. The litter of *A. nepalensis*, which is the dominant tree species, is known to produce nitrogen litter promoting soil carbon contents (Resh et al., 2002; Binkley., 2003). Soil Organic carbon is primarily derived from root exudates, plant residues, and litter and is influenced by forest species composition, forest history, and management (De Graaff et al., 2010). Litter biomass carbon and fine root biomass carbon were the main sources of soil carbon. Our study showed that litter biomass increased with stand basal area. Therefore, an increased nitrogen-rich litter amount leads to more soil organic carbon storage. Many studies have examined the effect of nitrogen-fixing tree species on forest soil (Binkley et al., 1992; Myrold et al., 1994; Resh et al., 2002; Binkley., 2003). Nitrogen-fixing tree species facilitate succession by increasing soil fertility, improving harsh environmental conditions, and possibly increasing soil organic carbon (Gomez-Aparicio et al., 2004; Macedo et al., 2008; Gomez-Aparicio 2009). Also, we found that soil organic contents and storage decreases along with soil depth. Our results revealed that soil carbon contents and storage were highest in the 0-30 cm soil depth and decreasing with soil depth and indicate the presence of organic matters on the upper soil layer. The soil organic carbon storage of 3.31-210.14 Mg ha<sup>-1</sup>

in the present study is within the average range of 121 to 344 Mg ha<sup>-1</sup> reported for other forest ecosystems (Lal, 2004). In the central Himalaya, the upper layer (0-30 cm) soil organic carbon storage is higher (Sheikh et al., 2009). Similarly, a previous study showed that amount of soil organic carbon content and storage was higher in the upper soil layer (0-30 cm) than the lower soil layer (FAO 2017). This indicates that soil organic carbon accumulation was mainly in the upper surface in the early stages of *A. nepalensis* forest development; and in late stages, soil carbon accumulated in the deeper soil layer.

### 4.3. Ecosystem carbon storage change

Ecosystem carbon storage increased significantly with the stand basal area and was mainly affected by tree species composition (Hu et al., 2015). Nitrogen-fixing tree species can accumulate more carbon than non-nitrogen-fixing tree species due to their fast-growing capacity (Chaer et al., 2011; Hoogmoed et al., 2014). The biomass carbon storage of reached 286.61 Mg ha<sup>-1</sup> in AMOO stand. Similar or higher biomass carbon stock has been reported from other oak forests in central Himalaya (Sharma et al., 2011). Variation in ecosystem carbon storage determined by the soil organic carbon and biomass carbon variations in stands. Moreover, the contribution of biomass carbon storage (Tree, shrubs, herbs, and litter) to ecosystem carbon storage decreasing with the stand basal area. Soil is the second-largest carbon storage after tree biomass. Soil organic carbon storage was 3.31, 31.21, 75.47, 157.04, 159.43, and 210.13 Mg ha<sup>-1</sup> at the AER, APDF, ALR, AMOM, AMR, and AMOO stand respectively, representing 20.89 %, 17.0 %, 34.9 %, 40.2 %, 39.5 % and 42.0 % of total ecosystem carbon. In addition, nitrogen-fixing tree species have a significant impact on forest soil and increase the carbon content of the soil. The colonization of pioneer *A. nepalensis* tree has been improving tree seedling establishment by acting as ‘succession facilitators’ improving soil nutrient status through nitrogen-rich litter production and providing favorable microclimatic condition too late succession tree e.g. *Q. leucotrichophora*, *R. arboreum* in the different forest (Callaway, and Walker 1997; Walker, and Reddell., 2007; Frouz et al., 2015). In our study area, the nitrogen-fixing *A. nepalensis* was the dominant species in term of standing biomass in early succession stand (AER, APDF, and ALR), as well as in late succession stands (AMOO, AMR, and AMOO), and accounted for a large part of the biomass storage. One conceivable reason for its success could be its potential capacity for atmospheric nitrogen fixation, which could significantly increase the rate of accumulation of biomass (Temperton et al., 2003; Knoth et al., 2014; Uri et al., 2017; Brookshire et al., 2019).

### 4.4. Implications for management

Our results revealed that the ecosystem C pools in the above and below-ground parts of the tree, shrubs, herbs, litter, and soil were changed significantly during *A. nepalensis* forests stand development. Developing forests has had a significant impact on biotic and abiotic factors, such as plant species, soil physicochemical properties, microbial communities, and the quantity and quality of inputs and outputs of organic matter (Myrold et al., 1994; Susaeta et al., 2014; Uri et al., 2014; Uri et al., 2017; Taerøe et al., 2017). The distribution of C between the plants and the soil during the development of the forest stand depends on the species of trees and *A. nepalensis* is a nitrogen-fixing tree that can change soil carbon more than any other non-nitrogen fixing tree. (Binkley et al., 1992; Myrold et al., 1994). *A. nepalensis* has been shown to support carbon storage in temperate forests in central Himalaya. The ecosystem C storage came primarily from *A. nepalensis*, which were more abundant and had a higher basal area than *Q. leucotrichophora* and *R. arboreum*. Our results provide new qualitative insight into the role of *A. nepalensis* in the temperate forest C sink in the central Himalaya. The Himalayan range is among the most fragile and unstable mountain regions of the world. Forest degradation in the Himalayas is a major challenge for forest managers and policymakers. The interplanting of nitrogen-fixing *A. nepalensis* with non-nitrogen-fixing tree species (*Q. leucotrichophora* and *R. arboreum*) may enhance the growth of the non-nitrogen-fixing tree species by increasing nitrogen availability (Binkley et al., 1992; Myrold et al., 1994; Semwal et al., 2013) improving nutrient cycling and soil fertility (Binkley., 2003; Khan et al., 2007; Bissonnette et al., 2014) and increasing carbon sequestration rate and improving risk management. The observation that *Alnus* raising soil nitrogen levels and subsequently influence the growth of the neighbor’s non-nitrogen-fixing species (Anthony and Klaus 2004). Maintaining or including nitrogen-fixing species in mixed forests appears to be an option for

enhancing soil carbon sequestration rate (Resh et al., 2002). Previous study indicates that *Alnus* species contribute significantly to the supply of nitrogen in the forest ecosystem thus markedly benefits soil fertility. In addition to being an early successional species, *A. nepalensis* colonize the recently-disturbed area with low levels of nitrogen availability. The obtained results show that *A. nepalensis* may play the role of early successional species in the temperate forest in central Himalaya. High nitrogen-rich litter of *A. nepalensis* reduced requirement of nitrogen in the soil and thus increase the forest productivity, regulating C cycle and mitigate global climate change. In the central Himalaya, *A. nepalensis* is currently of little importance in plantation forestry, although the potential for future use of *A. nepalensis* in forestry seems great and the opportunities are diverse. Although *A. nepalensis* does offer an alternative to chemical application of nitrogen to the forest, this potential of *A. nepalensis* systems in forest management is much more. That knowledge can contribute to the sustainability of forest practices which can ensure the restore the degraded forest and maintain their capacity to provide other goods and services for the benefit of current and future generations.

## CONCLUSION

Our results revealed that the abundance of *A. nepalensis* affect the ecosystem carbon storage. We found that tree biomass in different stands AER, APDF, ALR, AMOM, AMR, and AMOO were 8.97, 51.41, 16.07, 53.74, 144.77, and 101.14 Mg ha<sup>-1</sup> respectively. Soil organic C (SOC) storage of soil in different soil depth in successional stages AER (0-10 cm), APDF (0-30 cm), ALR (0-100 cm), AMOM (0-100 cm), AMR (0-100 cm), and AMOO (0-100 cm) was 3.31, 31.21, 75.47, 157.04, 159.43 and 210.13 Mg ha<sup>-1</sup>, respectively, with SOC concentration decreasing with increasing soil depth. The ecosystem carbon storage in the stand developing stand averaged 15.85, 183,216.26, 390.32, 403.66, and 500.08 Mg ha<sup>-1</sup> in the AER, APDF, ALR, AMOM, AMR, and AMOO respectively. In addition, trees and soils were the two largest contributors to the total ecosystem carbon pool in all stands. The soil carbon content and storage were highly heterogeneous among different stands and soil depths. Species community structure and composition had a significant influence on the biomass carbon storage and soil organic carbon storage of the stands. Our results are useful in estimating the total ecosystem C stock value of *A. nepalensis* forests in the study area. The *A. nepalensis* play an important role in stand development and enhancing ecosystem carbon storage by ecological succession. The expansion of *A. nepalensis* in central Himalaya can play an important role in regional carbon budget and degraded forest protection. Long-term monitoring and research are required to further explore the role of *A. nepalensis* forest stands in central Himalaya.

## DECLARATION OF COMPETING 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.

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**Table 1.** Geographical and vegetation characteristics across six study sites in central Himalaya

Stands	Latitude (N)	Longitude (E)	Altitude (m)	Aspect (°)	Slope (°)	Main tree species
AER	30°31' 51.54"	79°06'14.17"	1540	NW	22	<i>A.nepalensis</i>
APDF	30°33'01.35"	79°06'25.19"	1446	NE	25	<i>A. nepalensis</i> , <i>Q. leucotrichophora</i> , <i>R. arb</i>
ALR	30°31'50.63"	79° 06'10.69"	1540	NW	27	<i>A. nepalensis</i> , <i>Q. leucotrichophora</i> , <i>R. an</i>
AMOM	30° 32'56.84"	79°07' 50.91"	1335	NE	21	<i>A. nepalensis</i> , <i>Q. leucotrichophora</i> , <i>L. ov</i>
AMR	30°31' 22.63"	79 °06' 40.60"	1609	NS	28	<i>A .nepalensis</i> , <i>Q. leucotrichophora</i> , <i>R. an</i>
AMOO	30°35'6.12"	79° .01'36.71"	1599	NE	24	<i>A. nepalensis</i> , <i>Q. leucotrichophora</i> , <i>Q. fl</i>

**Table 2.** Proportion (%) of total stems by girth class (DBH) across six study sites in central Himalaya. Values in mean and stander error of three different three plot (n=3)

Proportion	0-10 cm	10-20 cm	20-30 cm	30-40 cm	40-50 cm	>50 cm
AER	100 <sup>a</sup> ±0.2					
APDF	2.5 <sup>a</sup> ±0.1	27.5 <sup>a</sup> ±1.1	37.5 <sup>a</sup> ±0.9	32.5 <sup>ab</sup> ±1.2		
ALR	86.9 <sup>ab</sup> ±1.2	7.9 <sup>a</sup> ±0.2	5 <sup>b</sup> ±0.1			
AMOM	3.57 <sup>a</sup> ±0.5	55 <sup>a</sup> ±0.3	28.6 <sup>b</sup> ±0.7	5 <sup>a</sup> ±0.2	2.1 <sup>b</sup> ±0.4	5.7 <sup>ab</sup> ±0.1
AMR	4.61 <sup>b</sup> ±0.3	14.47 <sup>b</sup> ±1.3	27 <sup>a</sup> ±1.3	17.7 <sup>a</sup> ±1.3	25.6 <sup>ab</sup> ±1.2	10.5 <sup>b</sup> ±1.1
AMOO		10.4 <sup>a</sup> ±0.5	19.5 <sup>b</sup> ±1.1	16.88 <sup>ab</sup> ±1.2	29.9 <sup>b</sup> ±0.4	23.4 <sup>a</sup> ±1.2

Note: Means in a column followed by different lower-case letters are significantly different at P < 0.05 (one-way ANOVA and LSD test).

**Table 3.** Community characteristics and Ecosystem carbon polls across six study sites in central Himalaya (Values in mean and stander error of three different plot).

Tree species/ Stand	Above-ground Mg ha <sup>-1</sup>	Below-ground C Mg ha <sup>-1</sup>	Basal area m <sup>2</sup> ha <sup>-1</sup>	IVI
<b>AER</b>				
<i>A. nepalensis</i>	5.65c±1.23	3.32b±0.54	5.2a ±0.1	300±0
Herbs	0.54b±0.03	0.12a±0.02		
Shrubs	1.26a±0.03	0.823c±0.01		
Litter	0.32c±0.1			
SOC (0-10 cm)	3.31a±0.54			
Ecosystem carbon storage	15.85c±2.13			
<b>APDF</b>				
<i>A. nepalensis</i>	40.61c±5.95	10.79b±1.51	27.81ab±5.22	141.33a±15
<i>Q. leucotrichophora</i>	7.10c±2.37	2.85b±1.18	2.02ab±0.70	38.51a±7.84
<i>R. arboreum</i>	16.49a±1.85	5.3a±1.16	10.74a±1.33	66.22a±8.88
Associated species	10.82a±1.14	3.01b±0.38	2.19ab±0.60	52.69a±11.7

Tree species/ Stand	Above-ground Mg ha <sup>-1</sup>	Below-ground C Mg ha <sup>-1</sup>	Basal area m <sup>2</sup> ha <sup>-1</sup>	IVI
Sub-total tree layer	75.01a±8.17	21.68b±3.86	42.56a±3.93	
Herbs	0.98c±0.23	0.35b±0.14		
Shrubs	1.23c±0.21	0.58b±0.12		
Litter	1.32c±0.13			
SOC (0-30 cm)	31.21ab±3.12			
Ecosystem carbon storage	183a±8.23			
<b>ALR</b>				
<i>A. nepalensis</i>	12.51b±1.99	3.55b±0.94	7.86a±0.55	58.13ab±1.6
<i>Q. leucotrichophora</i>	44.63d±13.71	21.49bc±5.96	4.71a±1.11	73.22a±9.78
<i>R. arboreum</i>	14.06b±2.66	5.8±1b.65	4.30a±0.77	58.14ab±6.7
Associated species	34.48c±14.90	10.54d±4.02	7.35a±0.64	117.16a±11
Sub-total tree layer	105.68c±20.82	41.37d±8.14	24.19c±1	
Herbs	1.32b±0.23	0.78c±0.12		
Shrubs	1.56d±0.34	0.92a±0.21		
Litter	2.56b±0.98			
SOC (0-100 cm)	75.47a±4.23			
Ecosystem carbon storage	216.26±10.32			
<b>AMOM</b>				
<i>A. nepalensis</i>	44.08b±1.83	9.65a±0.98	28.32b±2.54	82.44a±4.37
<i>Q. leucotrichophora</i>	44.87b±2.04	15.90a±1.5	10.32b±0.72	51.73b±3.60
<i>R. arboreum</i>	19.22b±1.22	6.78a±0.49	10.62b±0.94	49.15c±2.42
Associated species	75.62a±6.02	23.43b±2.84	13.62a±1.50	112.46a±2.4
Sub-total tree layer	183.58a±28.98	54.75ab±11.23	62.89ab±2.56	
Herbs	0.76b±0.14	0.31a±0.12		
Shrubs	1.35c±0.21	0.45a±0.26		
Litter	3.15a±0.98			
SOC (0-100 cm)	157.04b±9.23			
Ecosystem carbon storage	390.32ab±10.23			
<b>AMR</b>				
<i>A. nepalensis</i>	111.7a±7.3	27.4b±2.9	58.7a±2.6	171.95a±10
<i>Q. leucotrichophora</i>	22.45a±3.14	5.5a±1.16	7.7b±2.9	20.26a±2.22
<i>R. arboreum</i>	34.57a±2.87	11.17a±0.9	20.63a±2.33	97.55a±11.6
Associated species	1.44b±0.55	0.03a±0.01	0.89b±0.33	10.23b±5.44
Sub-total tree layer	170.15b±16.14	44.08a±5.92	87.90a±8.09	
Herbs	1.26a±0.12	0.54b±0.02		
Shrubs	1.78a±0.32	0.78ab±0.02		
Litter	3.56b±0.78			
SOC (0-100 cm)	159.43c±8.92			
Ecosystem carbon storage	403.66a±5.6			
<b>AMOO</b>				
<i>A. nepalensis</i>	84.86a±18.70	16.28a±7.07	40.58a±1.45	110.35a±1.8
<i>Q. leucotrichophora</i>	107.79b±25.93	30.12ab±7.02	32.25ab±1.83	98.21a±1.81
<i>R. arboreum</i>	5.57a±2.1	1.76a±0.8	5.12a±0.88	21.58a±3.21
Associated species	33.37a±11.48	8.12ab±4.11	10.46a±0.78	69.38b±1.29
Sub-total tree layer	231.56b±33.6	55.6a±10.98	88.39ab±4.88	
Herbs layer	0.89ab±0.23	0.21a±0.12		
Shrubs layer	1.43a±0.12	0.49b±0.13		
Litter	3.34b±1.33			
SOC (0-100 cm)	210.13c±10.43			

Tree species/ Stand	Above-ground Mg ha <sup>-1</sup>	Below-ground C Mg ha <sup>-1</sup>	Basal area m <sup>2</sup> ha <sup>-1</sup>	IVI
Ecosystem carbon storage	500.08b±14.4			

Note: Means in a column followed by different lower-case letters are significantly different at  $P < 0.05$  (one-way ANOVA and LSD test).

**Table 4.** Carbon poll in tree components across six study sites in central Himalaya. Values in mean and stander error of three different plot size (n=3) for biomass carbon. ‘—’ indicates no data.

Sites/Variables	Bole	Branch	Twig	Foliage	Catkin	Stump root	Lateral root	Fine
AER								
<i>A. nepalensis</i>	2.45±1.33	0.75±0.93	2.15±1.2	0.15±1.2	-	2.07±0.3	1.73±0.2	0.72±0.1
APDF								
<i>A. nepalensis</i>	29.69±5	9.33±3.6	1.4±0.99	1.86±1.3	0.53±0.3	6.78±1.4	4.28±0.8	0.6±0.1
<i>Q. leucotrichophora</i>	8.43±3.2	5.64±3.21	2.83±1.6	0.94±0.4	-	5.55±2.0	1.2±0.85	0.22±0.1
<i>R. arboreum</i>	14.76±4.1	5.61±2.05	4.71±1.7	1.56±0.1	-	5.42±2.4	2.56±1	0.81±0.1
<i>P. pashia</i>	12.27±1.2	7.83±2.15	3.6±2.16	2.71±1.06	-	4.76±2.1	0.48±0.1	0.05±0.1
<i>L. ovalifolia</i>	0.67±0.46	0.53±0.01	0.24±0.0	0.07±0.01	-	0.32±0.1	0.03±0	0.01±0.1
ALR								
<i>A. nepalensis</i>	8.01±1.4	3.25±0.77	0.75±0.1	0.71±0.1	0.16±0.04	1.74±0.3	0.48±0.2	0.21±0.1
<i>Q. leucotrichophora</i>	18.64±2.9	12.64±2.8	7.59±2.8	4.19±2.1	-	6.78±1.4	4.28±0.8	0.6±0.1
<i>R. arboreum</i>	7.41±1	3.4±0.77	2.34±0.6	0.69±0.2	-	4.12±0.7	1.6±0.26	0.66±0.1
<i>L. ovalifolia</i>	0.43±0.2	0.19±0.08	0.04±0.0	0.03±0.0	-	0.16±0.0	0.01±0.0	0.00±0.1
<i>N. pallens</i>	9±4.4	5.49±3.5	3.29±2.0	1.66±1.	-	4.62±3.0	0.68±0.2	0.10±0.1
<i>P. pashia</i>	1.86±0.45	1.02±0.1	0.65±0.1	0.20±0.	-	0.85±0.3	0.04±0.0	1.35±0.1
<i>M. esculenta</i>	4.89±0.82	2.81±0.9	1.65±0.5	1±0.32	-	2.46±0.7	0.2±0.01	0.03±0.1
AMOM								
<i>A. nepalensis</i>	33.51±3.4	11.23±1.	1.23±0.4	1.32±0.	0.27±0.1	5.72±2.4	2.16±1	0.80±0.1
<i>L. ovalifolia</i>	2.44±0.33	1.87±0.6	1.23±0.1	0.31±0.1	-	1.64±0.3	0.12±0.0	0.08±0.1
<i>Q. leucotrichophora</i>	22.31±1.9	13.76±1.	5.24±0.7	2.46±0.2	-	13.85±1	2.52±0.3	0.32±0.1
<i>Q. floribunda</i>	8.94±1.22	5.86±1.0	5.01±1.0	1±0.2	-	2.67±0.3	1.53±0.2	0.62±0.1
<i>A. indica</i>	7.8±4.5	3.07±1.77	2.81±1.62	0.49±0.28	-	1.68±0.97	5.8±3.35	2.74±0.1
<i>R. arboreum</i>	11.19±1.2	4.03±1.14	3.73±0.27	1.01±0.12	-	4.64±0.36	1.72±0.144	0.74±0.1
<i>N. pallens</i>	0.46±0.14	0.42±0.34	0.19±0.043	0.12±0.01	-	0.27±0.05	0.02±0.007	-
<i>P. pashia</i>	4.45±0.77	1.98±0.06	0.86±0.06	0.56±0.06	-	1.29±0.29	0.32±0.13	-
<i>M. esculenta</i>	1.9±0.47	1.14±0.32	0.58±0.144	0.67±0.20	-	0.69±0.11	0.08±0.02	0.00±0.1
<i>V. cylindricum</i>	3.4±0.64	2.39±0.66	0.75±0.22	0.67±0.15	-	1.34±0.32	0.4±0.29	-
<i>L. umbrosa</i>	0.88±0.16	0.23±0.06	0.08±0.02	0.01±0.10	-	0.34±0.07	0.02±0.00	0.02±0.1
AMR								
<i>A. nepalensis</i>	82.97±10.88	27.4±5.89	2.27±1.56	2.27± 2.71	0.63±0.45	24.40±9.3	4.15±2.37	0.35±0.1
<i>Q. leucotrichophora</i>	10.82±8.15	6.29±3.15	1.55±2.15	0.75±0.57	-	4.10±3.15	0.67±0.2	0.04±0.1
<i>R. arboreum</i>	16.77±10.15	6.72±4.15	5.84±3.15	1.41±1.15	-	6.65±4.15	2.75±0.12	0.92±0.1
<i>P. pashia</i>	0.69±0.58	0.43±0.17	0.17±0.05	0.33±0.12	-	0.22±0.12	-	-
AMOO								
<i>A. nepalensis</i>	61.37±16.97	19.81±4.93	1.41±0.97	1.41±0.47	0.41±0.07	13.15±1.12	2.2±0.35	0.12±0.1
<i>Q. leucotrichophora</i>	56.77±7.09	33.7±7.09	9.66±7.09	4.59±2.37	-	25.40±9.3	4.35±2.37	0.35±0.1
<i>Q. floribunda</i>	10.56±1.66	6.45±2.84	1.56±0.50	0.63±0.31	-	1.82±0.54	0.93±0.30	0.31±0.1
<i>R. arboreum</i>	2.93±0.95	1.36±0.82	1.02±0.47	0.25±0.17	-	1.04±0.50	0.45±0.34	0.15±0.1
<i>P. pashia</i>	0.64±0.11	0.55±0.02	0.21±0.09	0.17±0.01	-	0.36±0.07	0.02±0.03	-
<i>J. regia</i>	2.57±1.07	0.38±0.04	0.19±0.02	0.04±0.01	-	0.66±0.11	0.36±0.01	0.14±0.1

<i>A. indica</i>	1.56±0.50	0.62±0.32	0.38±0.17	0.08±0.00	-	0.78±0.11	0.47±0.11	0.12
<i>L. ovalifolia</i>	1.64±0.78	0.87±0.16	0.55±0.22	0.23±0.17	-	0.70±0.11	0.03±0.02	0.02
<i>B. capitata</i>	1.02±0.21	0.64±0.11	0.28±0.06	0.17±0.01	-	0.48±0.04	0.05±0.03	-
<i>N. pallens</i>	1.45±0.13	0.86±0.32	0.26±0.06	0.19±0.05	-	0.29±0.06	0.22±0.1	0.07





