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Altitudinal variation on physiological attributes of *Cedrus deodara* (Roxb.) G. Don in North-Western Himalaya

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Abstract
The study on "Altitudinal variation on physiological attributes of *Cedrus deodara* (Roxb.) G. Don in north-western Himalaya" was taken under four altitude range of <1800 masl (L_1), 1800-2100 masl (L_2), 2100-2400 masl (L_3) and >2400 masl (L_4) of Himachal Pradesh. This study represents the change in climatic conditions of Himalayan region may modify species distributions and performance of individual may also become affected in response to these variations along an altitudinal gradient. This study revealed that the highest total nitrogen (0.52%) and total carbon (0.72%) was found at <1800 m (L_1) followed by L_2, L_3 and L_4, respectively. The total nitrogen and total carbon content was decreased with increasing altitudinal ranges. The maximum total phenol (2.07 mgGAE/gm) was found at >2400 m (L_4) followed by L_3 (1.49 mgGAE/gm), L_2 (1.34 mgGAE/gm) and L_1 (1.21 mgGAE/gm). The total phenol content was increased along with increasing altitudinal ranges just opposite trend to total nitrogen and carbon. The highest chlorophyll (2.61 µg/ml), total sugar (7.76%) and total starch (9.61%) content was found at 1800-2100 m (L_2) and the lowest content was observed in L_1 (3.45%) and (5.30%), respectively. This study also revealed that the total nitrogen and total carbon content was decreased with increasing altitudinal ranges just opposite to total Phenol content in seedlings. Whereas, chlorophyll, total sugar and starch content were found maximum at 1800-2100 m and minimum content was observed at lower altitude.

Keywords: Altitudinal gradients, climatic conditions, physiological attributes, *Cedrus deodara*

Introduction
Deodar is the strongest of the coniferous woods; it is commonly used in building, furniture and carpentry. The wood oil of deodar is used for making perfumes and soaps. The bark astringent is useful for fevers, diarrhea and dysentery (Anon, 1950) [3]. Coniferous forests are important natural resources to sustain life in the Himalayas. The role of these forests lies in the maintenance of biodiversity; watershed protections as well as supplying timber, non-wood forest products, grazing land and habitat for threaten taxa (Ahmed *et al.* 2006) [2]. *Cedrus deodara* (Roxb.) G. Don is ecological & economically important conifer tree species of north – western Himalaya. This species is distributed from Afganistan to Garhwal (India) with an altitudinal range of 1200 to 3000 m (Gamble, 1881; Brandis, 1906 and Champion and Seth, 1968) [10, 5, 6].

In mountainous region, although with many factors contribute for variation in plant biodiversity but altitude has much more effect in shaping and limiting plant communities (Chawla *et. al.* 2008) [7]. Altitude, aspect and slope are the three main topographic factors which control the distribution and patterns of vegetation in mountain areas (Titshall *et al.* 2000) [22]. Among these three factors, altitude and aspect are most important ones. Altitude along with aspect in many respects determines the microclimate and thus large-scale spatial distribution and vegetation pattern (Day and Monk, 1974) [9]. The altitudinal limit resulting from various combinations of physical and biological factors determines the species distribution pattern. In conifers the natural regeneration practically depends on the seeds which in turn depends on production, dispersal germination capacity and successful establishment of seedlings However, seeds contain a lot of variation.

Shifts in C and N at the intraspecific level are consistently observed over altitudinal gradients (Korner *et al*. 1988; Vitousek *et al*. 1990; Marshall and Zhang 1993; Sparks and Ehleringer 1997) [12, 23, 15, 20]. However, the mechanisms underlying this trend are unknown. Ecologists have reported correlations of stable isotopes of N and C with the various abiotic factors over altitudinal gradients.

Leaf morphological and physiological traits that vary with altitude. Levels of N in plant are positively correlated with altitude (Korner 1989; Sparks and Ehleringer, 1997) [13, 20]. Likewise leaf mass per unit area increases with altitude (Woodwad, 1986; Williams *et al*. 1995) [28, 29] and is often correlated with $\delta^{13}C$ (Vitousek *et al*. 1990; Korner *et al*., 1991) [23, 14].

In natural ecosystems, various species have different adaptabilities to environmental changes, thus presenting different eco-physiological characteristics (Bhat, 2014) [4]. *C. deodara* is economically and ecologically important tree species of north western Himalaya. Hitherto no study has been carried out to ascertain the impact of climate changes, which has stated to impact adversely Himalayan ecosystem.

Material and Method

The experimental sites were carried out in *Cedrus deodara* (Roxb.) G. Don growing regions of the Himachal Pradesh. The study site lies between 30º96"06' - 30°96"96' N latitude and 077º20"13' - 077º19"65' E longitudes with an elevation ranging from less than 1800 to more than 2400 m above mean sea level. The samples have been collected from four altitude range of <1800 masl (L_1), 1800-2100 masl (L_2), 2100-2400 masl (L_3) and >2400 masl (L_4). Needles of *Cedrus deodara* were collected from the selected sites during October and November. The sample of needles from both mother trees and seedlings were quickly rinsed with de-ionised water to remove dust particles. All needle samples were oven dried at 60–80 °C. The elemental composition of needle samples was analyzed by the standard methods.

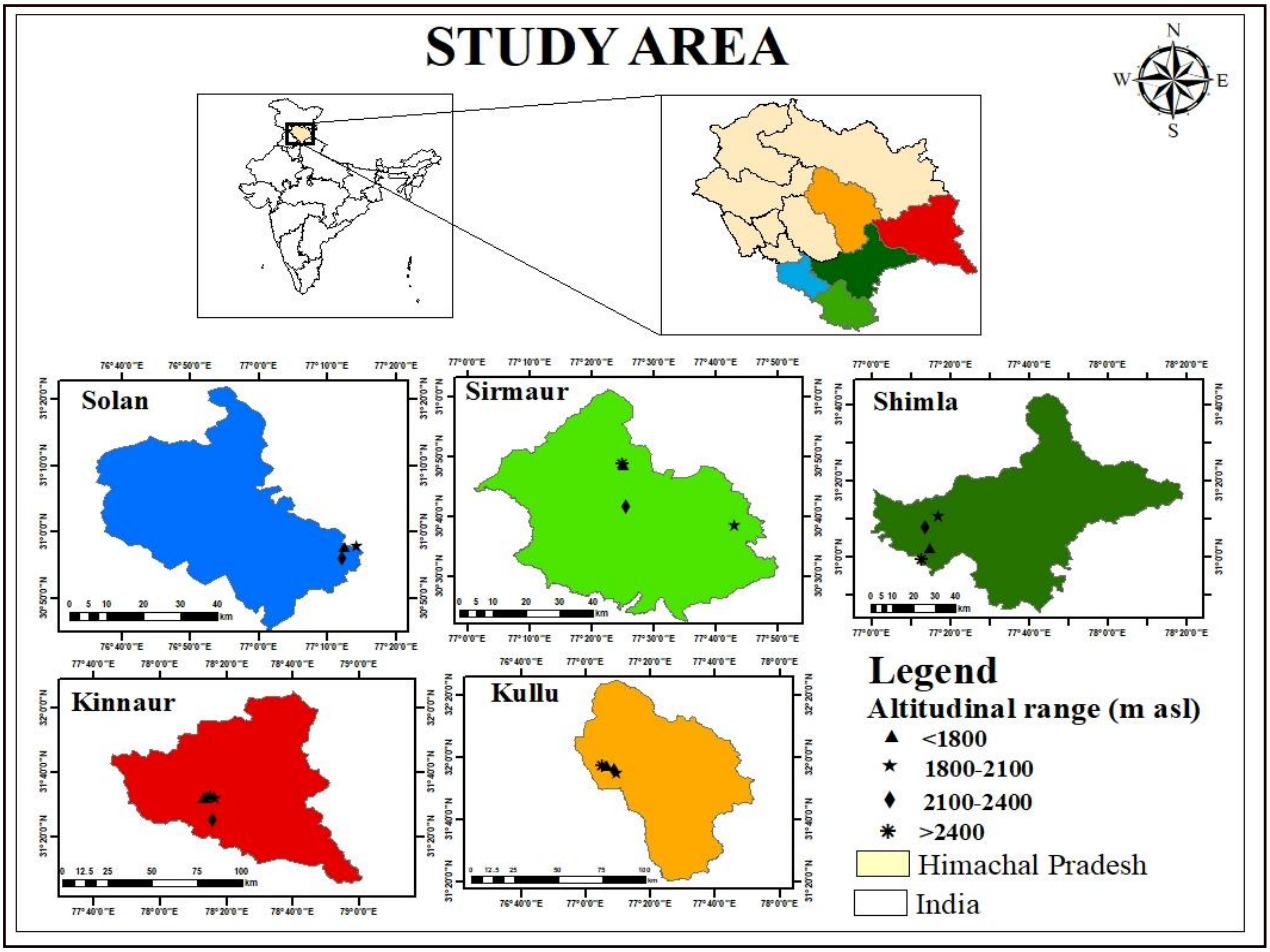

Fig 1: Map of Study Area

Results and Discussion

The data presented in Table 1 revealed that the highest total nitrogen (0.52%) and total carbon (0.72%) was found at <1800 m (L_1) followed by L_2, L_3 and L_4, respectively. The total nitrogen and total carbon content was decreased with increasing altitudinal ranges. Green needle nutrients also declined significantly (r^2 = 0.55) with increasing altitude. A similar trend of foliar nutrient concentrations was reported from several studies (Vitousek *et al*., 1988; Tanner, 1985) [24, 21]. The foliar nutrient concentrations would decline with increasing elevation (Tanner, 1985) [21], reflecting slower nutrient cycling as well as anticipated that foliar $\delta^{13}C$ would be less negative (^{13}C enrichment) and foliar $\delta^{15}N$ less positive (^{15}N depletion) at high altitudes. Hultine and Marshall (2000) [11] studied on the altitudinal trends in leaf nitrogen and stable carbon isotope composition in four conifer species: *Psedotsuga menziesii*, *Abies lasiocarpa*, *Picea engelmanni*, and *Pinus contorta*.

The maximum total phenol (2.07 mgGAE/gm) was found at >2400 m (L_4) followed by L_3 (1.49 mgGAE/gm), L_2 (1.34 mgGAE/gm) and L_1 (1.21 mgGAE/gm). The total phenol content was increased along with increasing altitudinal ranges just opposite trend to total nitrogen and carbon. The highest chlorophyll (2.61 µg/ml), total sugar (7.76%) and total starch (9.61%) content was found at 1800-2100 m (L_2) and the lowest content was observed in L_1 (3.45%) and (5.30%), respectively.

Table 1: Effect of altitudinal ranges on the biochemical attributes in needles of *C. deodara*

Altitude	Total Nitrogen (%)	Total Carbon (%)	Total Phenol (mgGAE/g)	Total Sugar (%)	Starch (%)	Chlorophyll (µg/ml)
<1800masl (L_1)	0.52	0.72	1.21	3.45	5.30	1.76
1800-2100masl (L_2)	0.41	0.67	1.34	7.76	9.61	2.61
2100-2400masl (L_3)	0.36	0.64	1.49	5.85	7.70	2.30
>2400masl (L_4)	0.27	0.59	2.07	4.69	6.54	1.88
SEm±	0.009	0.026	0.025	0.167	0.167	0.089
$CD_{(0.05)}$	0.018	0.053	0.050	0.336	0.336	0.179

Sims and Gamon (2002) + have reported that spectral indices provide relatively poor correlations with leaf chlorophyll content when applied across a wide range of species and plant functional types. Rao and Leblanc (1966) [16] reported that the reduction in chlorophyll content which is brought by acidic pollutants like SO_2. And CO_2-induced increase in phenolics was intensified by elevated temperature (Zvereva and Kozlov, 2006) [27]. In opposite, along altitudinal gradients, the total phenolics in *Quercus robur* foliage were found to decrease at lower elevations where temperature was warmer (Abdala-Roberts *et al*., 2016) [1].

Overall, the results of the present study supported that the altitudinal differences would lead to changes in physiological composition within a plant species. The observed differences in total nitrogen, carbon, total phenol, total sugar and starch and chlorophyll contents of needles across altitudinal gradients in our study are a result of the combination of environmentally induced variations in physiological differences where plants were exhibited.

Correlation Studies

1. Correlation coefficient (r) between altitudinal gradients (Alt) with biochemical contents of needles

Data presented in Table 2 reveals that the altitudinal gradient had significant negative correlation with total nitrogen (r = -0.270*). While, the altitudinal gradient had non-significant negative correlation with total carbon (r = -0.172) and total sugar & starch (r = -0.035). Altitudinal gradients depicted a non-significant positive relationship with total phenol (r = 0.198) it also revealed highly negative relationship with and chlorophyll (r = 0.027).

2. Correlation coefficient (r) between altitudinal gradients (Alt) with biochemical contents of seedlings

Altitudinal gradients (Alt) manifested a non-significant but negative relationship with total nitrogen (r = -0.195), total carbon (r = -0.220) and total soluble sugar and starch (r = -0.024) as well as with chlorophyll (r = -0.014). The correlation between altitudinal gradient and total phenol was observed to be non- significant but positive (Table 3).

Table 2: Correlation coefficients (r) between biochemical contents of needles under different altitudes

Traits	Alt	TN	TC	TP	TS	St
Alt	1.00					
TN	-0.270*	1.00				
TC	-0.172	0.505**	1.00			
TP	0.198	-0.660**	-0.431**	1.00		
TS	-0.035	-0.133	-0.057	0.081	1.00	
St	-0.035	-0.133	-0.057	0.081	1.000**	1.00
Ch	0.027	-0.051	0.105	-0.240*	0.669**	0.669**
Atl – Altitudes, TN - Total nitrogen, TC - Total carbon, TP - Total phenol, TS - Total sugar, St – Starch Ch –Chlorophyll					**Correlation is significant at the 0.01 level *Correlation is significant at the 0.05 level	

The correlation results in the present study found some support from the work done by earlier researchers. Hultine and Marshall (2000) [11] reported that the leaf nitrogen content and leaf mass per area were negatively correlated with carbon isotopes ($\delta^{13}C$). Sah and Brumme (2003) [17] studied Pine forest in Nepal in which the results revealed that the needle nitrogen content decreased about 0.57% over 800 m, from 1.67 N% at 1,200 m to 1.23 N% at 2,000 m and this negative correlation, however, was not found significant (–0.55). Carbon ($\delta^{13}C$) in the needles at the different altitudes ranged from –27.86 to –26.87% along with the altitudinal range of 800 m. A significant negative correlation (–0.87) was obtained between $\delta^{13}C$ abundance and altitude. Whereas stable carbon isotope compositions of C_3 plant species exhibited significant and positive correlation with the latitude observed (Zheng and Shangguan, 2007) [26]. They observed that the foliar $\delta^{13}C$ values of the plants in low altitude and high-altitude areas respond differently to elevation changes. In general, plants need a long period to adapt well to complex climatic environments. Increases in N could be associated with increased photo protection of the leaf against high light stress (Korner, 1989 and Sparks and Ehleringer, 1997) [13, 20]. However, changes on the nitrogen concentration on a per unit mass basis were small, indicating that the plant's nutritional status did not, necessarily, change very much on the gradient. There were, nevertheless, large changes in nitrogen in a per-area basis, mainly due to the changes in the specific leaf area, which could affect the functioning of the photosynthetic system.

Table 3: Correlation coefficients (r) between biochemical contents of seedlings under different altitudes

Traits	Alt	TN	TC	TP	TS	St	Ch
Alt	1.00						
TN	-0.195	1.00					
TC	-0.220	0.284*	1.00				
TP	0.213	-0.509**	-0.663**	1.00			
TS	-0.024	-0.364**	-0.033	0.169	1.00		
St	-0.024	-0.364**	-0.033	0.169	1.000**	1.00	
Ch	-0.014	-0.264*	-0.025	0.155	0.802**	0.802**	1.00
Atl – Altitudes, TN - Total nitrogen, TC - Total carbon, TP - Total phenol, TS - Total sugar, St – Starch Ch –Chlorophyll					**Correlation is significant at the 0.01 level *Correlation is significant at the 0.05 level		

Shi *et al*. (2012) [18] studied leaf N in different plant functional groups (including seed plants, fern, C_3 plants, herbs, woody plants, annual herbs, perennial herbs, and deciduous woody plants) collected from different altitudes ranged from 1200 to 4500 m in Tibetan Plateau, China. The general altitudinal pattern showed a significant and non-linear pattern of leaf N in plants with N_{mass} increasing below about 2200 m and then decreasing with increasing elevation above the altitude (R^2 = 0.243, p= 0.000) for most plant groups. Cui *et al*. (2018) [8] studied the effect of altitude on the physiological characteristics of *L*. *secalinus* on the Qinghai-Tibetan Plateau, where they measured the chlorophyll a (Chl *a*) and chlorophyll b (Chl *b*) in leaves from eight different altitudes from 1872 to 2935m asl. The Chl *a* and Chl *b* contents in HZ showed a significant negative correlation -0.971 and -0.968, respectively, with elevation.

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