



## RESPONSE OF PHENOLOGICAL EVENTS OF *Aesculus indica* Colebr. TO CLIMATE CHANGE ALONG AN ALTITUDINAL GRADIENT IN KUMAUN HIMALAYA, UTTARAKHAND

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### Abstract

The present study investigated the timing and duration of phenological events of *A. indica* in different elevational range in the Kumaun Himalayan forest. A total of four sites at elevation ranging between 1,900m and 2,200m were selected and at each site 10 trees were marked for observations. The phenological events, i.e. leaf bud formation, leaf bud busting, leafing, flowering bud formation, flowering bud busting, flowering, fruit/seed formation, seed fall and leaf fall were monitored. Phenological duration and asynchrony of these phenophases were determined at 10 day intervals and every 2-3 day intervals during the period of peak activities. The minimum length displayed leaf bud formation (44 days) and maximum by leaf fall (86 days) across the elevation. The environmental conditions, particularly temperature, affected the phenological patterns of *A. indica*. The leaf bud busting activity of *A. indica* was 51 days. Flowering activity started on May 1<sup>st</sup> and was extended over 76 days until July 15<sup>th</sup>. Seed fall activity was extended over 66 day across elevations. ANOVA showed the longevity of phenophases were varied significantly respective to elevations ( $p < 0.05$ ). Our observation showed that all the phenological events of *A. indica* appear early at lower elevations (1900 m) and are delayed with increasing elevation. All corresponding phenological events were earlier at lower elevations because the optimum temperature (9.0 to 19.5°C) is met earlier in these conditions.

Keywords: Elevation, Longevity, Phenological features, Temperature

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## Introduction

*Aesculus indica* (Colebr.) (English name: Indian Horse Chestnut, Family: Sapindaceae) is a medium to large deciduous, soft, perennial, temperate climatic plant abundantly found in the hills of Western and Central Himalayan forests (Sharma and Khanduri, 2007). Locally (in Kumaon) it is known as Pangar and occupies between 1,500 and 2,800m elevations in Kumaun Himalaya forests. It is distributed from Nepal northwestward into the State of Kashmir in north India, and across the Indus River to West Pakistan and to northeastern Afghanistan (Zhang *et al.*, 2010 and Syed *et al.*, 2016). In India, the tree is widely distributed in moist and shady ravines of Uttarakhand, Jammu and Kashmir, Himachal Pradesh and Uttar Pradesh (Singh, 2006; Zhang *et al.*, 2010 and Syed *et al.*, 2016). Zhang *et al.* (2010) also reported that the genus *Aesculus* is also found in eastern Asia, eastern North America, Europe and North Western Himalaya. The tree has high medicinal value, as oil from the seed is applied externally in the treatment of skin diseases and rheumatism. A paste is made from the oil cake is applied to the forehead to relieve headaches. The tree is being widely used for afforestation and ornamental purposes as well as for fodder, wood as small timber, and branches for making charcoal. In the forests, *Aesculus indica* seeds are eaten by monkeys, bears and other wild animals as fodder (Majeed *et al.*, 2009). The tree is commercially important and has high nutritional value as the fruits are fed to cattle and goats. The seed embryo is ground and mixed with flour, providing staple food for the hill people (Sharma and Khanduri, 2007). At lower elevation, it mixes with *Pinus roxburghii* and other broadleaf species, while at higher elevation it remains associated with *Quercus leucotrichophora* and *Quercus floribunda* in the forest. The subtropical forests are located along an altitudinal gradient and exhibit limited day length variation within the annual cycle. However, temperature, particularly at higher elevation, approaches those of temperate latitudes. Phenological observations provide a background for information on functional rhythms of plants and their communities (Singh *et al.*, 2015).

Phenology, in its simplest term, is the study of events of nature in response to seasonal and climatic changes to the environment (Singh, 2014). It involves precise documentation of timing and duration of different phenological events at species level, their interrelations and possible causal links between environmental variables (Singh and Kushwaha, 2005). The knowledge of the phenological patterns of fruit tree crops is essential to estimate their behavior in a new growing area by extrapolation of experimental results from one region to another, or to predict changes in the tree phenology in a given area caused by the variation of the environmental factors between years (Broadhead *et al.*, 2003). Thus, phenological studies can provide criteria for selection of suitable genotypes, helping to improve fruit yield and quality, and reduce environmental risk (Petri *et al.*, 2008). If the phenology of a species shifts at a rate different from that of the species that constitute its ecological conditions, it will lead to variation in its seasonal activities (Visser *et al.*, 2004) lead to a mismatch in phenology (Stenseth and Mysterud, 2002). The occurrence of phenophases is determined by the

local biotic climatic characteristics of the plant species (Leon-Ruiz *et al.*, 2011). In plants, bud-burst, leaf-expansion, abscission, flowering, fertilization, seed set, fruiting, seed dispersal and germination, all take place in respective seasons. The phenology of plant communities can be studied by dealing with particular life-history stages separately, such as leafing, flowering, fruiting, seed dispersal and germination. Each of these events occurs in its own calendar slot, but there is clearly interdependence between them. Fruiting must wait upon flowering; seed dispersal cannot proceed fruiting. Each phenomenon can be studied at different levels of organization.

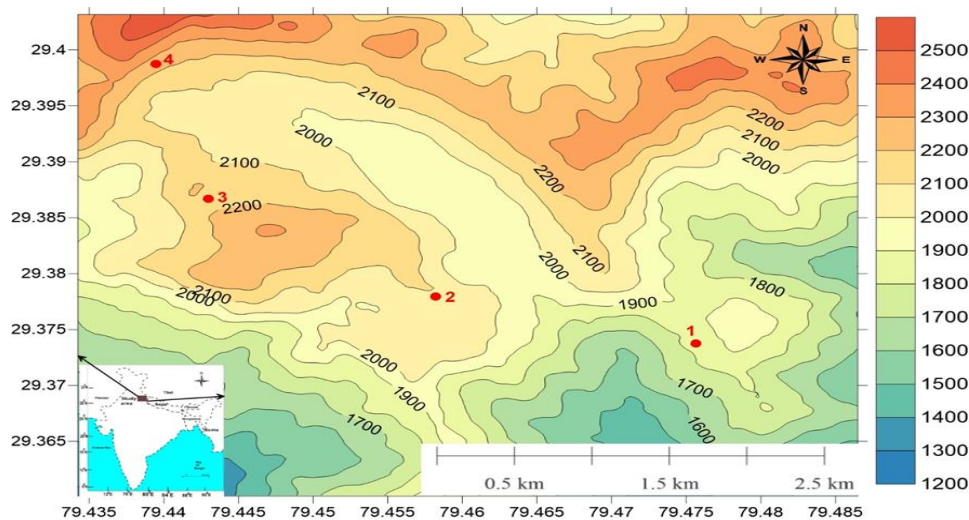
Climate indicators such as variation in temperature and rainfall, closely influence phenophases (Thakur *et al.*, 2008) and variation in timing, duration and synchronization of phenological events of different tree species (Borogayary *et al.*, 2018). It is evident that climate changes will occur during the long lifespan of tree species and changes in phenology may be the major visible short-term response (Badeck *et al.*, 2004). In fact, tree phenological observations have proven to be the most effective impact indicators of climate change (Kushwaha and Singh, 2008). Climate change effects on seasonal activity in terrestrial ecosystems are significant and well documented, especially in the middle and higher latitudes (Badeck *et al.*, 2004). Ground observations of phenology not only bear a clear and consistent warming signal, but also indicate parallelism in the phases of warming and advancement of phenology (Badeck *et al.*, 2004). The impact of climate change on the plants can be correlated with respect to the changes in the flowering time. Most species react differently to climate change; the outstanding question is how future climate change will affect the phenology of the whole ecosystem under changing climate scenarios. Climate is probably the most important determinant of vegetation patterns globally and has a significant influence on the distribution, structure and ecology of forests (Singh *et al.*, 2015). In the recent climatic conditions many plants species changed their vegetative as well as reproductive phenophases. In some locations the phenophases appear early or in other locations it delayed with the same species. *Aesculus indica* are globally distributed from lower to higher elevation in varying climatic conditions in different parts of the world. The record of phenological events of *A. indica* in recent climatic condition provides noticeable information in future research. Thus, the present study has been undertaken to focus on the documentation of the phenological events of *Aesculus indica*, and to correlate it with the effect of climate change with particular reference to elevation gradients.

## **Materials and Methods**

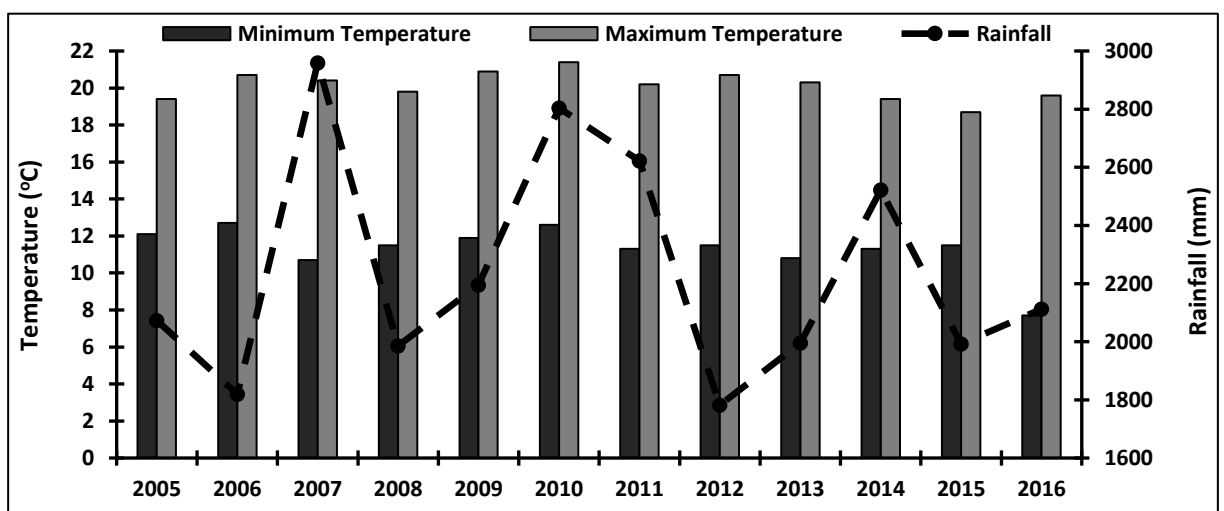
The study was conducted in the Kumaun Himalaya area, North India, approximately 305 km North of Delhi. The vegetation in this area is dominated by *Shorea robusta*, in the sub mountain zones (up to 1000 m), and *Quercus leucotrichophora*, in the low-to-mid mountain zones (above 1700). In between these zones, *Pinus roxburghii* forest is dominant. The associated species with *Q. leucotrichophora* are viz. *Quercus floribunda*,

*Rhododendron arboreum*, *Aesculus indica* and *Myrica esculenta*. Four plots were selected at every 100m elevational between 1900 and 2200m a.s.l. (29°22'– 29°23' N; 79°26'–79°27' E) in locations with eastern aspect (Figure 1 and Table. 1). Soil types in the region are dominated by arenosols and cambisols.

Overall climate at the studied elevations ranges from subtropical to temperate type, with pronounced differences between seasons. Winter months (December to January) are usually very cold, with small amounts of rain and heavy snowfall, and summers (April to mid-June) are warm and dry. A warm and humid rainy season occurs from mid-June to mid-September. Along 1,900 to 2,200m elevation, the total annual precipitation was 1,992mm and mean average varied from 11.5 to 18.7°C. January is the coldest month (3.9°C) and May the warmest (26.39°C) (Figure 2). The rainfall and temperature data were taken from Aryabhatt Research Institute of Observational Science (ARIES), Nainital during the study period.



**Figure 1.** Location map of the studied sites and plots in different elevation.



**Figure 2.** Variation in rainfall and temperature data in the study sites over ten years

Phenological observations were made on 10 trees selected 10x10 m per plot. Tree selection was based on the following criteria: healthy trees approaching maturity, with well-developed crowns and free from buttresses. The main morphometric variables, tree height and circumference at 1.37m, were measured for all trees, using a Ravi multimeter for height (patent no. A 000191, India) and a measuring tape for circumference. Overall, the circumference of *A. indica* trees varied between 39.8 and 48.6cm, with a mean circumference of 44.6cm ( $\pm 0.27$ ), and tree height varied between 12.2 and 19.1m, with a mean height of 15.18m ( $\pm 1.94$ ). The mean values of morphometric variables (tree height and circumference at 130cm) for the trees at each site are given in table 1.

**Table 1.** Site location and main morphometric data (height and circumference) of the sampled *Aesculus indica* trees at each plot. Values within brackets are standard errors.

Site	Elevation (m)	Coordinates	Number of sampled trees	Average height (m)	Average circumference (m)	Associated Species with <i>A. indica</i>
1	1900	29°22'38"N 79°28'30"E	10	16.53 ( $\pm 0.66$ )	0.442 ( $\pm 0.007$ )	<i>P. roxburghii</i> , <i>M. esculenta</i> , <i>R. arboreum</i> ,
2	2000	29°23'05"N 79°27'30"E	10	15.81 ( $\pm 0.47$ )	0.469 ( $\pm 0.005$ )	<i>Q. floribunda</i> , <i>R. arboreum</i> , <i>C. macrophylla</i> ,
3	2100	29°23'23"N 79°26'39"E	10	14.29 ( $\pm 0.52$ )	0.451 ( $\pm 0.009$ )	<i>Q. leucotrichophora</i> , <i>Q. floribunda</i> , <i>R. arboreum</i> , <i>L. ovalifolia</i> , <i>C. torulosa</i>
4	2200	29°23'55"N 79°26'33"E	10	14.09 ( $\pm 0.49$ )	0.421 ( $\pm 0.007$ )	<i>Q. leucotrichophora</i> , <i>R. arboreum</i> , <i>L. ovalifolia</i> , <i>C. viminea</i> , <i>M. duthiei</i>

Phenological observations were made throughout a period of one year, from January 2015 to January 2016. For each event, observations were made at every 2-3 days during peak activity periods and every 10 days during low activity periods (Singh *et al.*, 2015). Every phenophase was considered to be at peak when more than 70% of the marked tree showed the phenological event and when no phenological events were observed it was considered to be at a low activity period. On each monitoring day, the presence or absence of a certain phenological event was recorded for each selected tree. An event was considered to be active just when it was observed in at least 20% of the crown of a tree, otherwise it was noted as absent. Assessment of the crowns was performed by direct observation from the ground.

### **Data analysis**

For statistical comparisons among the different elevations, the dates of beginning and end of each phenological phase were converted into days of the year. All comparisons were performed using the nonparametric Kruskal-Wallis tests, since the assumption of normality was frequently violated. A multiple comparison post-hoc test (Kruskal-Wallis test), using two-tailed hypothesis testing, was then applied to assess the significant differences between each pair of sites. Analysis was performed with IBM SPSS Statistics software (IBM SPSS Statistics V. 23, IBM Corporation, Armonk, NY, USA) at a significance level of 5% ( $\alpha = 0.05$ ). Significance was ascribed for  $P$ -value less than 0.05. Levels of significance are indicated by the number of symbols (\*\* $P \leq 0.05$ ; \* $P \leq 0.01$ ; \*\*\* $P < 0.001$ ). Data are presented as mean  $\pm$  SD.

### **Results and discussion**

The results obtained from various phenological events, i.e. leaf bud formation, leaf bud busting, leafing, flowering bud formation, flowering bud busting, flowering, fruit formation, seed fall and leaf fall were monitored and studied (Figure 3).

In *A. indica* at Site 1, leaf bud formation, leaf bud busting and leafing started from February 25<sup>th</sup>, March 10<sup>th</sup> and March 15<sup>th</sup> respectively, while at Site 4 it was delayed by 18, 20 and 16 days respectively. The period of leaf bud formation was 34, 31, 26 and 36 days (Figure 3) across the site, it was earlier at Site 1 (25<sup>th</sup> February) and late in Site 4 (15<sup>th</sup> March). The mean temperature during the starting time of leaf bud formation was 11.0, 7.5, 10.0 and 9.25<sup>o</sup>C (Figure 4) across the site. The period of leaf bud busting was 32, 26, 26 and 32 days, site 1-4 and 2-3 showed the similar longevity of phenophases but different in timing, it was earlier started in site 1 (10<sup>th</sup> March) and delayed in site 4 (30<sup>th</sup> March) (Figure 3). The mean temperature during the starting time of leaf bud busting was 10.0, 9.25, 13.75 and 10.75<sup>o</sup>C (Figure 4) from site 1 to 4. The timing of leaf initiation was from 15<sup>th</sup> March to 15<sup>th</sup> May across the sites. The period of leafing from site 1 to 4 was 42, 36, 36 and 45 days, site 4 showed the longest leafing period. (Figure 3). During leaf initiation, the mean temperature was 9.25, 11.75, 12.75 and 12.75<sup>o</sup>C (Figure 4) from low to high elevation. Yadav and Bisht (2013) have reported

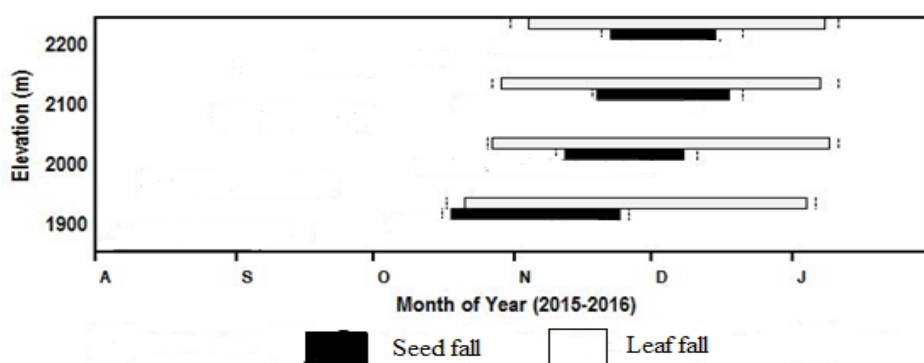
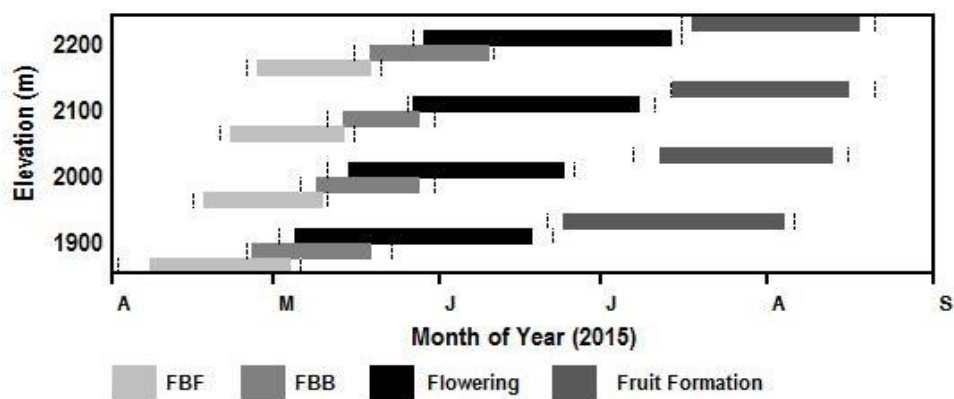
the leaf flushing period in *A. indica* during the mid of March, the leafing activity was 5 – 10 days earlier in the current observation in comparison to the previously reported timing. At low elevation site, leaf bud formation, leaf bud busting and leafing occur in comparatively low mean temperature, while in higher elevation it appears with increasing mean temperature.

At site 1, first flowering bud formation was observed on April 5<sup>th</sup>, flower bud busting was observed on April 25<sup>th</sup> and first flowering was observed on May 1<sup>st</sup>, while flowering bud formation and flowering bud busting occurred after 20 day and flowering after 30 day at site 4. The end of the flowering was observed on June 20<sup>th</sup> at 1900m, 25<sup>th</sup> June at 2000m, 10<sup>th</sup> July at 2100m and 15<sup>th</sup> July at 2200m elevation in the present study. The duration of flowering bud formation was 30 and 26 days. Site 1 showed the maximum time period of flowering bud formation, while the other three sites showed the same period of 26 days (Figure 3). During flowering bud formation the mean temperature in the selected site was 9.0, 14.25, 15.0 and 18.25<sup>o</sup>C (Figure 4). The time of flowering bud formation occurred on 5<sup>th</sup> April to 25<sup>th</sup> April across the sites. The flowering bud busting was 25<sup>th</sup> April to 15<sup>th</sup> May across the sites, the duration of flowering bud busting was 25-25 days at site 1 and 2, whereas 20 and 26 days at site 3 and 4 (Figure 3). Flowering bud busting started when the temperature was 18.25, 19.50, 21.75 and 19.75<sup>o</sup>C (Figure 4). The first flowering period was 1<sup>st</sup> May at site 1, 10<sup>th</sup> May at site 2, 25<sup>th</sup> May at site 3 and 30<sup>th</sup> May at site 4. The duration of flowering in different sites/elevation was 51, 46, 47 and 46 days. Site 1 showed slightly more duration compared to other sites (Figure 3). Bud busting or flowering need an optimum temperature, soil moisture, rainfall, humidity and wind speed to initiate the phenological events. In the present study the mean temperature during flowering for selected study site in different elevations was 16.25, 21.75, 24.50 and 25.00<sup>o</sup>C (Figure 4). We have compared the flowering activity period of *A. indica* with an earlier study by Sharma and Khanduri (2007), which reported that the first floral bud was observed on 7<sup>th</sup> April at 2,050m elevation and on 11<sup>th</sup> April at 2,550m elevation, flowering was reported on 13<sup>th</sup> April. Borchert (1994) reported flowering in *A. indica* in May – June at 1,500 – 2,800m elevation. Sharma and Khanduri (2007) reported the end of flowering at both the elevation (2050 and 2550m) from the 26 – 27<sup>th</sup> May. The flowering bud formation was shown at an earlier timing and flowering was at a delayed timing as compared to the earlier study.

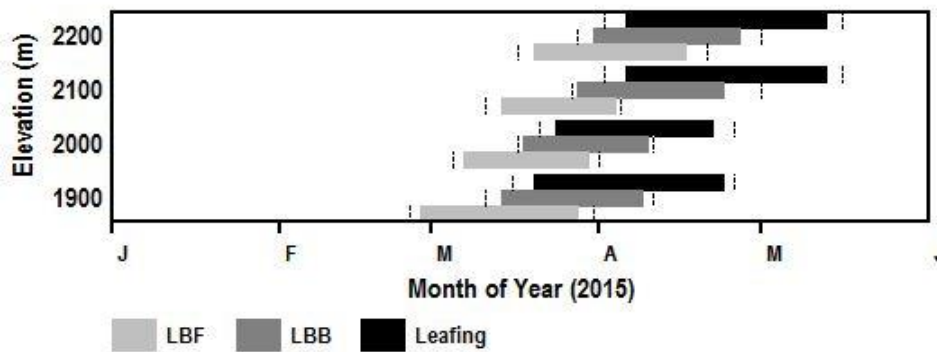
The fruit formation in *A. indica* started on 20<sup>th</sup> June to 5<sup>th</sup> August at site 1, while it was after 25 and 35 days at site 4. The seed fall activity occurred after 35 days in the higher elevation when comparing seed activity to the lower elevation. The seed fall ending time was on 25<sup>th</sup> November at site 1 and 20<sup>th</sup> December at site 4, the seed fall period was 25 days earlier at lower elevation site. The duration of fruit formation was 47 days at site 1, 41-41 days at site 2 and 3, and 36 days at site 4. The longevity of fruit formation was comparatively low at site 4 (Figure 3). The mean temperature during fruit formation was 19.5, 20.9, 17.6 and 21.75<sup>o</sup>C (Figure 4) across the sites. The seed fall was started on 15<sup>th</sup> October at site 1 and it was delayed by 25, 30 and 35 days at

site 2, 3 and 4 respectively from site 1. The duration of seed fall was 46, 30, 35 and 31 days from site 1 to site 4 (Figure 3). During the starting of seed fall, the mean temperature was 15.05, 14.65, 15.6 and 13.25°C (Figure 4) across the sites. Fruit setting in *A. indica* reported by Sharma and Khanduri (2007) in the last week of May to the first week of June. Fruit setting timing was delayed in the present observation compared to previous reported timing.

The first leaf fall timing observed in *A. indica* was on 15<sup>th</sup> October at site 1 and 1<sup>st</sup> November at site 4 and it ended on 5<sup>th</sup> January at site 1 and 10<sup>th</sup> January at site 4. The time of leaf fall was after 15 days but the end timing was after 5 days only at the lower to the higher elevation. The timing of leaf fall was earlier at the lower elevation and delayed with increasing elevations. The longevity of leaf fall was 82, 76, 71-71 days from site 1 to site 4 (Figure 3). During leaf fall starting time the mean temperature was 15.05, 14.95, 13.8 and 14.9°C (Figure 4) across the sites. The starting and ending of leaf fall period in earlier study by Sharma and Khanduri (2007) was from the end of October to the first week of November and was completed by the end of November. Yadav and Bisht (2013) reported that the leaf drop period in *A. indica* was between the second week of October to the last week of November.





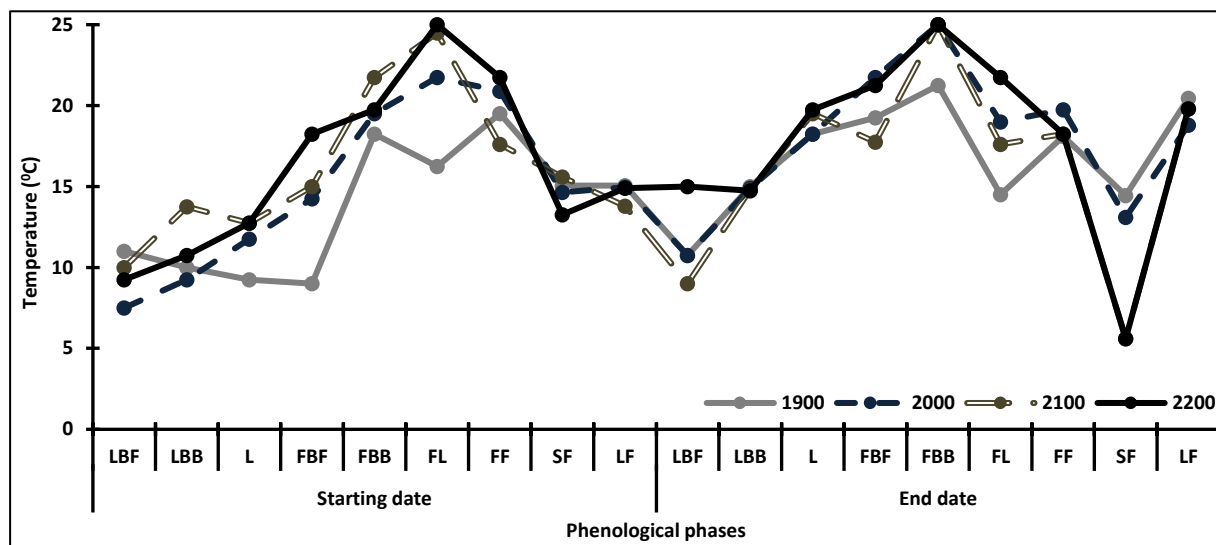


**Figure 3.** Timing and duration of phenological phases of *A. indica* along the studied elevation gradient. Phases represented are: Leaf Bud Formation (LBF, light grey); Leaf Bud Burst (LBB, dark grey) and Leafing (black) (upper panel); Flower Bud Formation (FBF, light grey), Flower Bud Burst (FBB, intermediate grey), Flowering (black) and Fruit Formation (dark grey) (central panel); Seed Fall (black) and Leaf Fall (white) (lower panel). Each phase was recorded as present when it could be observed in at least 20% of the buds/shoots of a tree. Shaded areas represent the average duration of each phase at each elevation.

Tree phenological observations have proved to be the most effective impact indicators of climate change (Chmielewski and Rotzer, 2001; Kushwaha and Singh, 2008; Moza and Bhatnagar, 2005). The species in some ecosystems are so strongly adapted to the long-prevailing climatic pattern that these are vulnerable even to modest changes (Luo *et al.*, 2007). Global climate change is likely to alter the phenological patterns of plants due to the controlling effects of climate on plant ontogeny (Singh *et al.*, 2010). Changes in the timing of seasonally re-occurring biological events/phenology are one of the most powerful biological responses to environmental change. Climate change in the Himalayan region is critical because it will impact not only the environment of the mountains themselves but also the large and highly populated areas adjacent in the plains (Ralhan *et al.*, 1985).

Winter temperature and soil moisture was the most important variable affecting the regression model for phenophases at all sites. The result showed the phenological events of *A. indica* in relation to varying elevations. The variations in phenological events of *A. indica* were due to the changes in climatic condition like, temperature, soil moisture, humidity and rainfall which varied from one site to another (Singh *et al.*, 2015). The warmer temperature immediately following the low temperature may aid in this process by inducing bud bursting (Negi, 2006). The concentrated leafdrop activity immediately followed the sprout of leafing in evergreen tree species and leafing and flowering is a simultaneous process after bud bursting (Singh *et al.*, 2015). In the present study the starting time of flowering in *A. indica* was observed when the mean temperature was highest, while in the ending time flowering bud busting showed the maximum mean

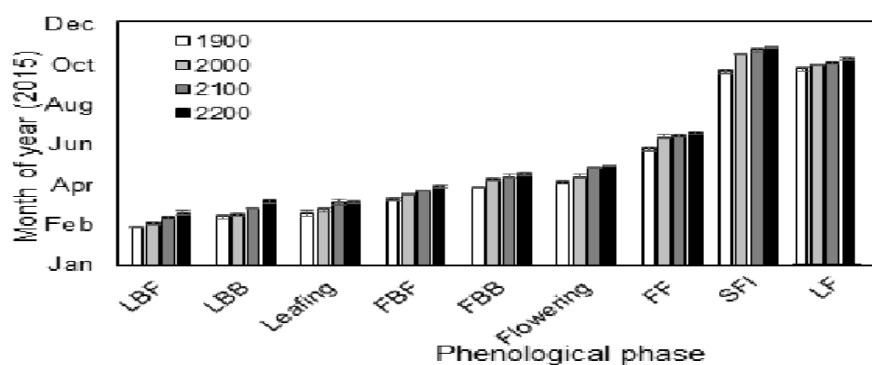
temperature. The mean minimum temperature was observed during initiation time of leaf bud formation across the sites (Figure 4)



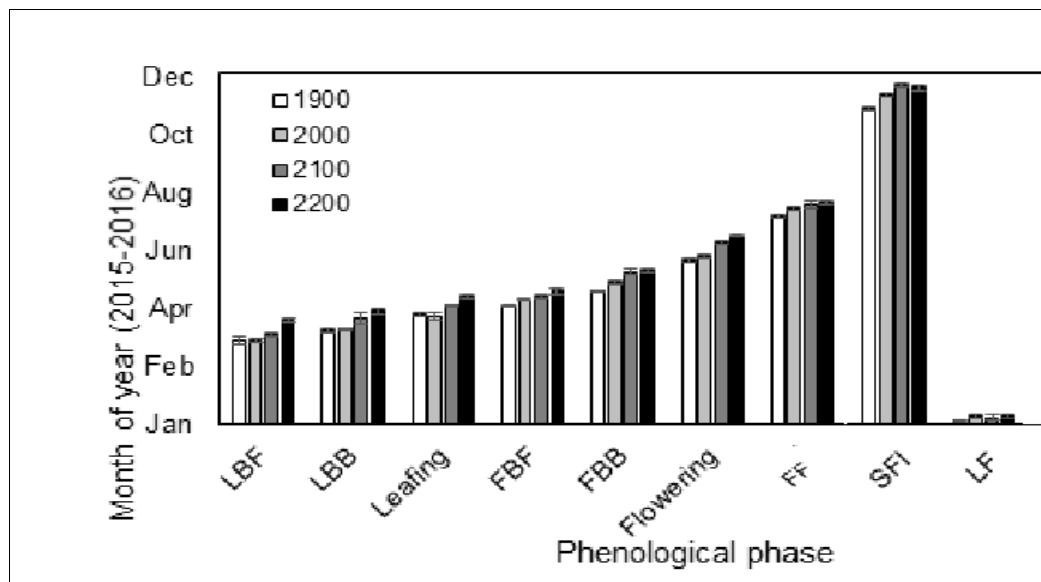
**Figure 4.** Variation in temperature during starting and ending timing of phenological events at selected studied elevational gradient.

The variations in phenological timing of *A. indica* were due to the changes in climatic condition because temperature, soil moisture, humidity, rainfall, water stress and wind speed which varied from elevation to elevation. Global temperatures have increased by up to 0.6°C over the past century, and with rising temperature and enhanced evapo-transpiration rate, ecosystems may suffer from severe water deficit over the next few years (Tewari *et al.*, 2018). When the temperature increases during early summer, it induces the growth of phenophase, similarly when the temperature drops during early winters, termination is triggered. The leaves fall, fruit drops and the seed becomes dormant as a result of the sudden increase and drop of temperature (Sharma and Khanduri, 2007). In the present study all the phenological events like, leaf bud formation, leaf bud busting, leafing, flowering bud formation, flowering bud busting, flowering, fruit formation, seed fall and leaf fall occurred earlier at lower elevation and were delayed at increasing elevation (Figure 5 & 6). Phenological activities are additionally inhibited by moderate water deficits (Ashman and Schoen, 1997). Occurrence of leaf flushing (vegetative phase) and flowering (reproductive phase) requires the availability of substantial amounts of resources within the trees. For instance, flower production and maintenance requires considerable expense of energy to form non-photosynthetic tissues and nectar (Singh and Kushwaha, 2006; Omondi *et al.*, 2016). Various physiological activities or sinks (e.g. leaf buds and leaves, flower buds and flowers, and fruit) may compete for water, nutrients and metabolites (Singh and Kushwaha, 2006), and such internal competition may lead to the partitioning in timing of plant functions like leafing and flowering. While the two survival adaptations (leafless period and flowering time) are linked with capacity

adaptation (e.g. resource-use rate), the duration of deciduousness and time lag between the onset of leafing and flowering helps trees in making the maximum use of the available resources like water, soil minerals and nutrients for growth and reproduction (Singh and Kushwaha, 2006). The leafless period is an adaptation to avoid water stress, and water stress affects flowering time in tropical forest trees (Shaver, 1981). The phenological events at lower elevation sites appeared comparatively with low temperature, while the same phenological events at higher elevation sites appeared slightly with high temperature. All the phenological events were earlier at the lower elevation because the optimum temperature was met earlier at the lower elevation and met later with increasing elevation. At the lower elevation, all the phenological events, leaf bud formation, leaf bud burst, leafing, flower bud formation, flower bud burst, flowering, fruit formation, seed fall and leaf fall were observed earlier by two to three weeks due to the micro climatic variation and long photo-period.



**Figure 5.** Variation of phenophase starting dates among the elevational gradient. Different letters represent significant differences between sites. Error bars are Standard deviations.



**Figure 6.** Variation of phenophase end dates among the elevational gradient. Different letters represent significant differences between sites. Error bars are Standard deviations.

The longevity of leaf bud formation, leaf bud busting and leafing was longer at higher elevation, flowering bud formation, flowering bud busting and flowering longevity did not show any significant change along the elevation and fruit formation, seed fall and leaf fall longevity were shorter with increasing elevation. Leaf longevity is important both as a nutrient cycling process and as a specific plant adaptation (Kumar *et al.*, 2013). Generally, the longevity of phenophase decrease with increasing elevation which may be due to the low temperature, low water stress and low soil moisture at higher elevation. Soil temperature showed a significant correlation with flowering phenology (Dahlgren *et al.*, 2007; Singh, 2014). Dahlgren *et al.* (2007) reported that many plant taxa show a direct relationship between leaf drop and water stress. Due to lower rate of water stress at higher elevation sites, leaf fall showed a shorter period comparison to lower elevation sites. The minimum longevity was shown by leaf bud formation (44 days) and maximum by leaf fall (86 days) across the elevation in *A. indica*. Leaf bud formation, leaf bud busting, seed fall and leaf fall activity occurred in winter months when the temperature was low, while the leafing, flowering bud formation, flowering bud busting and flowering activity occurred in summer months when the temperature was high and the rest of the phenological events occurred in the spring months.

ANOVA showed that the longevity of leaf bud formation ( $F = 62.75$ ,  $p < 0.05$ ), leaf bud busting ( $F = 20.75$ ,  $p < 0.05$ ), leafing ( $F = 64.75$ ,  $p < 0.05$ ), flowering bud formation ( $F = 18.75$ ,  $p < 0.05$ ), flowering bud busting ( $F = 22.00$ ,  $p < 0.05$ ), flowering ( $F = 24.75$ ,  $p < 0.05$ ), fruit formation ( $F = 40.75$ ,  $p < 0.05$ ), seed fall ( $F = 82.00$ ,  $p < 0.05$ ) and leaf fall ( $F = 50.75$ ,  $p < 0.05$ ) varied significantly with elevations (Table 2).

**Table 2.** Analysis of variance for longevity of different phenophases across selected elevations

Dependent Variable	Source	Type III Sum of Square	DF	F
Elevation	Leaf bud formation	188.25	3	62.75*
	Leaf bud busting	62.25		20.75*
	Leafing	194.24		64.75*
	Flowering bud formation	56.25		18.75*
	Flowering bud busting	66.00		22.00*
	Flowering	74.25		24.75*
	Fruit formation	122.25		40.75*

	Seed fall	246.00		82.00*
	Leaf fall	152.25		50.75*

Note: R Square = 0.950 (Adjusted R Square = 0.931) and \* Significant at 0.05%

The correlation between longevity of fruit formation of *A. indica* and elevation showed that it was negatively significant with leaf bud formation, leafing and flowering bud busting. The longevity of seed fall and flowering bud busting showed negative correlation with the elevation. Similarly, the longevity of leaf fall showed the negative correlation with leaf bud formation and leafing in the elevation. While all other phenological events of *A. indica* were positively significant with each other phenophases ( $P < 0.01$  and  $P < 0.05$ ) (Table 3).

**Table 3.** Correlation between longevity of different phenophases and elevations.

	<b>LBF</b>	<b>LBB</b>	<b>L</b>	<b>FBF</b>	<b>FBB</b>	<b>FL</b>	<b>FF</b>	<b>SFI</b>	<b>LF</b>
<b>LBF</b>	1.00	0.896**	0.965**	0.430	0.830**	0.321	-0.195	0.085	-0.041
<b>LBB</b>		1.00	0.866**	0.733**	0.652*	0.668*	0.154	0.487	0.280
<b>L</b>			1.00	0.300	0.727**	0.207	-0.353	0.009	-0.218
<b>FBF</b>				1.00	0.334	0.987**	0.784**	0.885**	0.855**
<b>FBB</b>					1.00	0.179	-0.102	-0.139	0.046
<b>FL</b>						1.00	0.824**	0.948**	0.873**
<b>FF</b>							1.00	0.836**	0.985**
<b>SFI</b>								1.00	0.843**
<b>LF</b>									1.00

Note: \*\*. Correlation is significant at the 0.01 level (2-tailed) and \* Correlation is significant at the 0.05 level (2-tailed). Leaf Bud Formation (LBF); Leaf Bud Burst (LBB); Leafing (L); Flower Bud Formation (FBF); Flower Bud Burst (FBB); Flowering (FL); Fruit Formation (FF); Seed Fall (SFI) and Leaf Fall (LF).

## Conclusion

To summarise, it can be concluded that the phenological events of investigated tree species appeared earlier at the lower elevation and was delayed with increasing elevation. Changes in temperature affected phenological events of *A. indica* to a greater degree at higher elevations. The magnitude of the response of

phenological events of *A. indica* to temperature variations was significantly correlated along the elevational gradient. Besides temperature as a key variable in this study, other irregularities in phenological events of *A. indica* are dependent in other abiotic factors. There could be many other factors such as water stress, rainfall pattern, soil moisture, wind speed, nutrients and oxygen levels that would be useful to better understand spatial patterns in the sensitivity of phenological responses to temperature. Hence, more detailed investigations at the local level are required to examine the influence of these events in future studies.

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