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Leaf Phenological Diversity of Dominant Trees of Sub-Tropical Forest of Mizoram, Northeast India: Effect of Environment and Species Intrinsic Quality

R. Lalruatfela, S. K. Tripathi

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ABSTRACT

Variations in leaf phenophases (length of deciduousness) of tropical forest trees are widely reported as a result of adaptive mechanisms of different species to harsh environmental conditions as reflected by tree intrinsic quality. Various leaf stages (leaf flush, maturation and senescence) of 20 tree species were monitored periodically in sub-tropical forest of Mizoram. Patterns of leaf flush and leaf fall were correlated with tree intrinsic quality parameters, e.g. stem wood density (SWD), wood moisture content (WMC), leaf mass area (LMA), specific leaf area (SLA) and leaf strategy index (LSI). In the studied forest, leaf flushing was most prevalent during the wet months except in some species where asynchronous pattern (regardless of seasons) of leaf flush was observed which was more prevalent in leaf exchanging species. LSI was significantly positively correlated ($p < 0.01$) with length of deciduousness, for example, LSI was maximum (0.7) in >2 months deciduous species followed by 0.5–0.7 in < 2 months deciduous species and 0.3–0.5 in leaf exchangers. The LMA was negatively correlated with LMC. In 90% of the species, the months of maximum SLA coincide with the months of minimum LMA. The LMC was significantly positively correlated with the wood moisture content, whereas, SLA and LMC was significantly negatively correlated ($p<0.01$) reflecting an increase in plant water use efficiency with increase in SLA. This study indicated a strong role of mild climatic variability on leaf phenophases in sub-tropical forests. Study suggested that tree phenological information can be used as a bio indicator to climate change response, particularly changes in precipitation and temperature patterns.

Keywords Phenological diversity, Abiotic variables, Tree intrinsic quality, Leaf strategy index, Leaf mass area.

R. Lalruatfela, S. K. Tripathi*
Department of Forestry, Mizoram University, Aizawl 796004, India
e-mail: sk_tripathi@rediffmail.com
*Corresponding author

INTRODUCTION

The term phenology was derived from the Greek word *Phiano* meaning to show or to appear. It is a science of recurring events in nature characterized by various developmental stages of leaf e.g. leaf initiation, maturity and senescence. Phenology was defined as the study of the timing of recurrent biological events, the causes of their timing with regard to biotic and abiotic forces and the interrelation among phases of the same or different species (Lieth 1974). The life cycle events of most organisms are reported to be relatively fixed in time, which are often strongly determined by climatic factors (i.e. temperature and precipitation / water availability). Therefore, changes in these environmental conditions are reported to strongly affect many phases in the life cycles of most plants (Kushwaha et al. 2010, Kushwaha et al. 2011a), for example, the timing of leaf-burst, defoliation, flowering, seed-setting and ripening (Kushwaha et al. 2011a and b). Changes in phenological patterns are the outcome of environmental conditions experienced by the species within an ecosystem, which significantly affect the competitive ability of species in the ecosystem. The species responses to climate will depend on individual species within its area of distribution and therefore, changes in phenological behaviors of the species will differ according to the site (Kushwaha et al. 2011a and b). Therefore, understanding the relationship between the various response factors and climate would be important for selecting suitable bio-indicators to detect and monitor the landscape-ecological effects of climate change on plants.

Plant phenology has been considered as one of the most responsive and easily detectable traits in nature that changes in response to climate. Changing patterns of plant developmental stages brought about by the current anthropogenic global climate change can considerably affect plant productivity, inter-specific plant competition and interactions with heterotrophic organisms (Tripathi 2010). Such changes may considerably affect the capacity of the ecosystem to provide goods and services to the society which needs to be monitored through setting up long term ecological research station (Tripathi 2010). Major climatic factors (e.g. temperature and precipitation) have been reported to determine seasonality in phenology of plant leaf and roots (Tripathi et al. 1999, Marques et al. 2004, Tripathi et al. 2005, Wapongnungsang and Tripathi 2019) as a result of changes in tree water status (Reich 1995), which is necessary step for predicting the impacts of anthropogenic climate change on plants and their ecosystems (Lalnunzira and Tripathi 2018) by affecting their flowering and fruiting patterns in relation to environment (Singh and Kushwaha 2006, Boulter et al. 2006).

Among the tree intrinsic quality parameters, leaf mass area (LMA) has been reported as one of the important factors to significantly affect inter - specific variation in relative growth rate (Garnier 1992) which affect leaf life span both positively as well as negatively (reciprocal to deciduousness) and photosynthetic rate per mass, respectively (Wright et al. 2004). Further, LMA along with stem wood density (SWD) and leaf strategy index (LSI, resource use or photosynthesis rate) have been found to significantly affect deciduousness and leaf longevity in dry tropical trees (Kushwaha et al. 2011a and b). Therefore, LMA of a species can be used as a good indicator of resource acquisition of the species. Evergreen species have been reported to have higher LMA than deciduous species (Sobrado 1991, Villar and Merino 2001) however, differences between families are larger compared to evergreen and deciduous species. Specific leaf area (SLA), an important indicator of leaf area index (Xiao et al. 2006), has been reported to be positively correlated to relative growth rate (Poorter et al. 2009) and site productivity. SLA depends on light conditions and the longevity of leaves (Gholz et al. 1976, Del Rio and Berg 1979), which differs between evergreen and deciduous trees (Gower and Richards 1990, Withington et al. 2006). Phenological studies are important for the management of forests in changing climate. Wide variations in the length of deciduousness are reported for the dry tropical tree species (Kushwaha et al. 2010) as a result of strong seasonal variations in the climatic events (i.e. rainfall and temperature).

In tropical moist forest, variations in the length of deciduousness are not presented to be so high because of prolonged rainy season and a short dry period where the studies are limited. This study reports changes in vegetative phenology (changes in leaf phenophases) of important trees in tropical rain forest and find out possible environmental cues for changes in sub-tropical tree phenophases. Further, we will explore the relationship between climatic variables, leafing phenology and various tree parameters to find out the possible environmental and tree intrinsic causes to leaf phenology.

This study addresses the following major questions in sub-tropical broad leaved forest of Mizoram: (1) Whether leaf phenological diversity exists among the tree of sub-tropical forests ? (2) How does leaf phenological pattern of different tree species changes with seasons ? (3) How do critical tree intrinsic factors affect leaf phenological patterns in this region ?

MATERIALS AND METHODS

Study site

The present study was conducted in Hmuifang reserve forest (23º.45´ N and 92º.7´ E) about 50 km apart from Aizawl district of Mizoram, Northeast India. The altitude of forest varied from 1500–1619 m a.m.s.l. The mountain is still covered with virgin forests reserved since the Mizo Chief's time. Hmuifang forest is ideal to the nature lovers and one of the major source of water for Tuirial river. The vegetation of Hmuifang forest belongs to Northern Sub-Tropical Broad Leaved Hill Forest type (subgroup 8-B of Champion and Seth 1968). The surrounding forests near Hmuifang Mountain abound with fascinating varieties of flora and fauna including rare orchids and wildlife especially avifauna. The topography is undulating but the slopes of the hills are gentle. Trees growing at the edge of the hills on the windward side show stunted growth as well as those that are growing in more open spaces. The reason for sparse undergrowth and fairly moist ground can be attributed to the trees out of which majority of them are leaf exchanging species and sunlight barely reaches the ground vegetation. The study is conducted for two annual cycles i.e. May 2016 to April 2018.

Temperature and Rainfall data were collected from Mizoram Remote Sensing Application Center, Science and Technology (MIRSAC) New Capital Complex Road, Khatla (Fig. 1). The climate is characterized by three seasons namely summer (March-June), rainy (July-October) and winter (November–February). The mean monthly maximum (day) temperature varies from 25ºC in January to 34ºC in May and the mean minimum (night) temperature from 11ºC in January to 24ºC in May. Long-term annual rainfall varies between 2140 and 2563 mm. The study was carried out during two annual cycles (i.e. from May 2016-April 2018). The total rainfall and mean temperature during the 1st cycle were 2167 mm and mean temperature was 18.95ºC, respectively. The corresponding values for rainfall and temperature during the 2nd annual cycle were 2564 mm and 20.85ºC. The peak rainfall was recorded in June and August during the first and second annual cycles (Fig. 1).

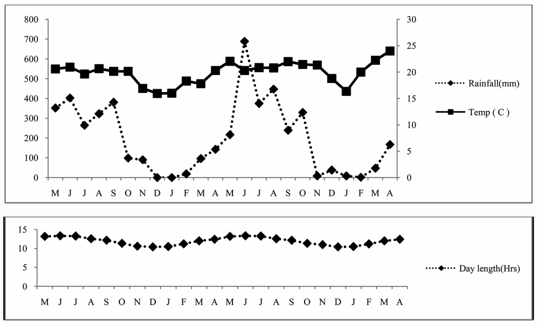

Fig. 1. Upper diagram represents total monthly rainfall (left Y axis) and mean monthly temperature (right Y axis). Lower diagram represent monthly day length.

Experimental design

Phenological observations were recorded in 20 selected trees species at maturity with girth at breast height (GBH) ≥ 30 cm. For each species, a total of 5 individual trees were marked with spray paint and this way a total of 100 tree species were marked for periodical observation. On each tree, four major branches, each facing different direction were selected and four twigs (currently growing shoots of last-order branches) on each of four major branches (one in each direction) were marked with metal tags. Phenological events were recorded at monthly observation between May 2016 and April 2018. Leaf phenological events were recorded for the timing and duration of leaf flush and leaf fall, monthly leaf moisture content (LMC), monthly leaf weight and monthly leaf area. In brief, we considered leaf exchanging species if the deciduousness period was <10 days, brevi-deciduous species if the leaf less period was 10-30 days and deciduous species if the leaflessness was 30 days and above. Further, leaf mass area (ratio of leaf mass to leaf area g^{-2}), leaf strategy index (ratio of leaf fall duration to leaf flush duration), specific leaf area (ratio of leaf area to leaf weight cm^2g^{-1}), wood moisture content (WMC) and stem wood density (SWD) were calculated. Each month 20 leaves from each tree species were collected with a total of 240 leaves per species per year and a total of 4800 leaves measured per year for all species marked. Leaf size classes (microphyll–2.25 to 20.25 cm^2, mesophyll–20.25 to 182.25 cm^2 and macrophyll–182.25 to 1640.25 cm^2)were distinguished as per the classification proposed by Raunkiaer (1934). Leaves were dried

Table 1. Tree intrinsic quality parameters (e.g. stem wood density, SWD, leaf mass area, LMA and leaf strategy index, LSI) and length of deciduousness of important tree species of Hmuifang sub-tropical forest of Mizoram, Northeast India. Values are mean ± 1SE.

Species name	SWD (g cm –3)	LMA (g m–2)	LSI	Deciduousness (Days)
Leaf exchanging				
Diospyros lanceifolia	0.643 ± 0.022	97.182 ± 4.73	0.422 ± 0.0017	6 ± 2
Lithocarpus xylocarpus	0.673 ± 0.014	87.879 ± 4.7	0.411 ± 0.0041	8 ± 2
Symplocos racemosa	0.719 ± 0.020	87.919 ± 6.37	0.484 ± 0.0011	7 ± 3
Symplocos theaefolia	0.669 ± 0.038	95.366 ± 6.78	0.385 ± 0.0011	6 ± 2
Syzigium claviforum	0.839 ± 0.020	104.846 ± 9.03	0.487 ± 0.0018	9 ± 3
Brevi-deciduous				
Belschmeidia roxburghiana	0.545 ± 0.020	95.417 ± 6.32	0.481 ± 0.0031	10 ± 2
Callophylum polyanthum	0.604 ± 0.056	88.732 ± 5.41	0.525 ± 0.0022	11 ± 2
Ixora walichii	0.715 ± 0.011	73.096 ± 2.96	0.473 ± 0.0021	13 ± 3
Quercus spicata	0.59 ± 0.011	83.041 ± 5.14	0.506 ± 0.0026	14 ± 2
Thing chhunghul	0.721 ± 0.036	60.262 ± 6.31	0.532 ± 0.0021	14 ± 2
Schima walichii	0.689 ± 0.012	64.728 ± 2.19	0.569 ± 0.0015	20 ± 5
Dipterocarpus retusus	0.666 ± 0.016	98.871 ± 6.96	0.657 ± 0.0069	26 ± 2
Ostodes paniculata	0.424 ± 0.012	46.79 ± 4.86	0.54 ± 0.0033	27 ± 4
<2 months deciduous				
Eriobotrya bengalensis	0.783 ± 0.047	110.671 ± 8.75	0.702 ± 0.0036	58 ± 2
Helicia excelsa	0.685 ± 0.041	87.716 ± 6.15	0.678 ± 0.0010	33 ± 4
Rapanea capitellata	0.59 ± 0.031	91.285 ± 8.94	0.641 ± 0.0033	39 ± 3
2–3 months deciduous				
Styrax polysperma	0.546 ± 0.022	77.871 ± 2.94	0.769 ± 0.0095	75 ± 4
Embelia tsjeriam	0.771 ± 0.014	93.762 ± 6.54	0.852 ± 0.0031	88 ± 3
Quercus floribunda	0.793 ± 0.059	89.932 ± 7.86	0.743 ± 0.0033	63 ± 5
Engelhardtia spicata	0.511 ± 0.005	67.077 ± 4.67	0.726 ± 0.0015	65 ± 7

in an oven at 70ºC for 48 h to measure the moisture content, leaf mass area and specific leaf area. Leaf area was measured manually using a graph sheet. Wood samples were dried at 105 ºC in an oven for 48 h, percent wood moisture content was measured using the formula (wmc = initial weight–oven dry weight / oven dry weight × 100). Statistical analysis was done using Microsoft excel and SPSS software.

RESULTS AND DISCUSSION

Periodical phenological analysis of 20 marked trees indicate that 5 tree species belong to leaf exchanging/ semi/-evergreen category and 8 species belonging to brevi-deciduous category and remaining species were exhibiting varying degrees of deciduousness (Table 1). Majority of leaf exchanging species exhibit leaf flush between April and May after the onset of rain when the soil moisture improved and few species produce leaf during the winter between October and February during dry period.

Among the deciduous species, majority of them exhibit leaf flush between April and May and the rest during the end of monsoon or during winter. For a majority of the species except a few, leaf exchange occurs almost throughout the year, besides, asynchronous leaf flush is also seen in some species. Based on the majority of the observation, there is less pronounced seasonality in the majority of the tree species and leaf flush is almost absent during the peak months of rainfall i.e. June and August. The timing of senescence is observed mainly at the end of monsoon and during the winter season. For the

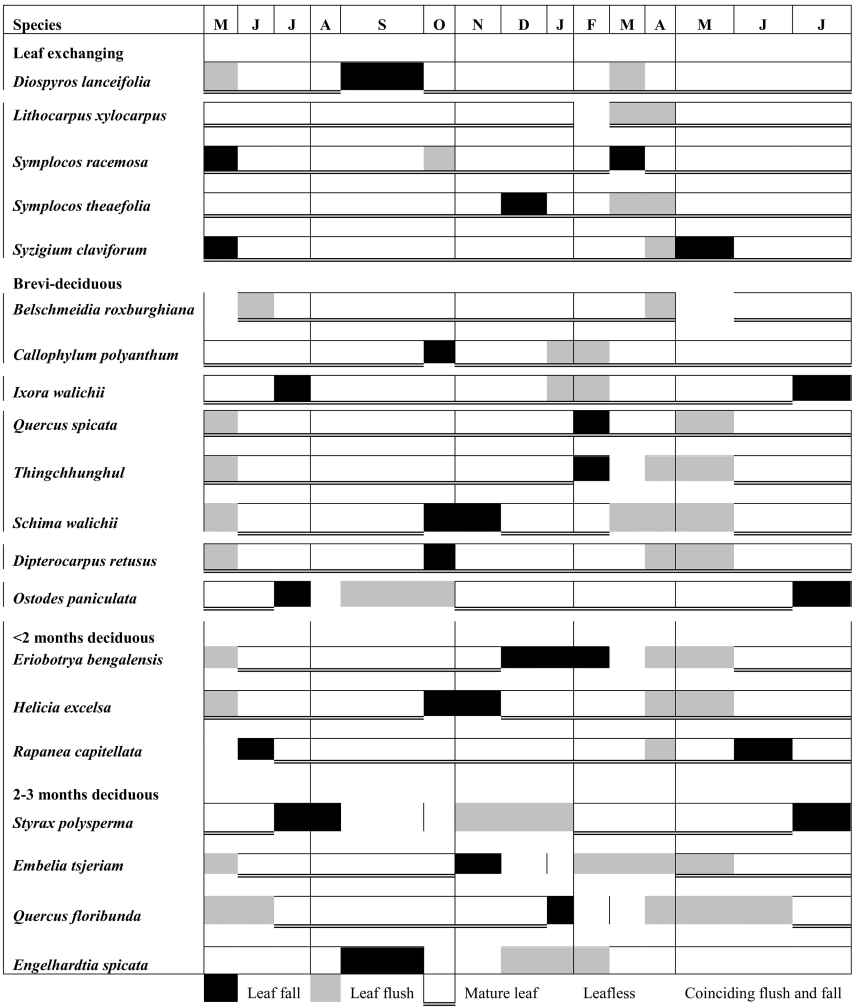

Fig. 2. Variation in different leaf phonological patterns in selected trees of sub-tropical forest of Mizoram, Northeast India.

majority of the species, phenological activity is low during the monsoon, which can be attributed to the absence of prolonged dry season and the availability of moisture throughout the year (Kushwaha et al. 2011a and b).

Most of the deciduous species (viz. *Quercus floribunda, Dipterocarpus retusus, Styrax polysperma, Eriobotrya bengalensis, Engelhardtia spicata*, *Embelia tsjeriam* and *Helicia excelsa*) in the present study exhibited leaf flush in the beginning of the growing season which stayed until autumn. This phenomenon of leaf growth was corresponded with the increase in LMA during leaf maturity. Earlier observed similar trend in Northern Hardwood forest of America which explained that it may be because of the buildup of cell wall material and chloroplast. However, in leaf exchanging species (i.e. *Callophylum polyanthum* and *Ixora walichii*) the LMA drops at the time of leaf exchange and the leaf resources are quickly replenished by the time of leaf maturity. In some species (e. g. *Syzigium claviforum*, *Rapanea capitellata*, *Eriobotrya bengalensis, Ostodes paniculata* and *Helicia excelsa*) LMA increases considerably (>60%) from the monthly average values which was coincided with leaf fall and leaf nutrient retranslocation during senescence. Tree species with low LMA tend to have a high concentration of proteins and minerals, high water content, low concentration of lignin and other secondary compounds and a fast metabolism (high rates of photosynthesis and respiration per unit leaf dry mass) (Lambers and Poorter 1992, Wright and Westoby 2002) (Fig. 2).

The length of deciduousness (days) was significantly positively correlated (r = 0.94, p < 0.01, n=20) with LSI. The relationship between leaf area and WMC was significantly positive (r = 0.57, p < 0.01, n=20) and the correlation between LMC and SWD was significantly negative (r = –0.405, p < 0.01, n=20). We found that months of maximum leaf moisture content (LMC) coincided with months of leaf flush. Further, it is also found that the maximum monthly LMA values were negatively correlated (r = –0.453, p < 0.05, n = 20) to monthly leaf moisture content. The SWD and LMA were positively correlated (r = 0.53, p < 0.01, n = 20). The LMC was significantly positively correlated with WMC (r = 0.63, p<0.01, n=20). WMC and SWD were significantly negatively correlated with each other (r=–0.49, p<0.05, n=20). WMC was species specific, among the trees observed, *Quercus floribunda* has the minimum WMC (45.5%) whereas *Ostodes paniculata* has the maximum WMC (142%).

SLA increases with decreasing light conditions, and thus with canopy depth. Stand structure has a similar influence on the light conditions and therefore also affects SLA (Del Rio and Berg 1979, Tardieu et al. 1999, Nagel and O'Hara 2001, Poorter et al. 2009), which holds true for *Ostodes paniculata* and *Thing chhunghul* ; a tree of lower height among the species studied. It is found that in the entire deciduous tree species observed, the month of maximum SLA coincides with the month of leaf production. It is also found that in 90% of the species, the months of maximum SLA always coincide with the months of minimum LMA and vice versa (r = –0.911, p<0.01, n=20), meaning that there is a reciprocity between LMA and SLA (Kushwaha et al. 2011a).

SLA was significantly positively correlated (r = 0.529, p < 0.05, n=20) with SWD and significantly negatively correlated (r = 0.529, p < 0.05, n = 20) with LMC. This indicates that decreasing SLA value in tree leaves increases water use efficiency of the species. Further, SLA was significantly negatively correlated with WMC (r =–0.454, p < 0.05, n = 20) and LMA (r = –0.826, p < 0.01, n=20). Majority of the species exhibited minimum value of monthly SLA in the transition period between winter and summer (i.e. February to May) during the dry months with lowest precipitation indicating plant strategy to hold water in the plant tissues to avoid water stress for their metabolic activities.

Raunkaiers classification of leaf size (cm^2): Microphyll–2.25 to 20.25, mesophyll–20.25 to 182.25 and macrophyll–182.25 to 1640.25, LA–leaf area, SLA-specific leaf area, LMC–leaf moisture content.

Monthly average temperature and rainfall are strong variables regulating the tree leaf flushing in tropical montane sub-tropical forest, for example, majority of the tree species exhibited leaf flushing when the monthly average temperature crosses 20^{o}C and the monthly average rainfall exceeds 100 mm. However, moisture stress cannot be suggested as a criterion for senescence in sub-tropical montane forest which is presumably related to the genetic makeup of the species (Table 2).

Out of the trees that are studied *Eriobotrya bengalensis, Rapanea capitellata*, *Dipterocarpus retusus, Helicia excelsa* and *Ostodes paniculata* are seen to remain deciduous for a short interval spanning not more than two months while *Quercus floribunda*, *Engelhardtia spicata*, *Embelia tsjeriam* and *Styrax ploysperma* remain deciduous for more than 2 months and the remaining are leaf exchanging species. The longest deciduousness was shown by *Embeliats jeriam* spanning more than three months. In leaf exchanging species, marked differences in leaf flush can be obtained even within the same species, in majority of the species, senescence starts earlier in branches that bear greater amount of fruit. Early leafing is observed during the II annual cycle owing to the increase in precipitation. Irrespective of being deciduous or evergreen, double leaf flush per annual cycle is also observed. It was observed that overall leaf density and leaf size for the same species was lesser on higher elevation and foliage development

Table 2. Leaf characteristics (evergreen and deciduous), classification and leaf are (LA), specific leaf area (SLA) and leaf moisture content (LMC) of selected trees of subtropical forest of Mizoram, northeast India.

Species name	Family	Leaf persistence	Raunkiaers classification	LA (cm^2)	SLA (cm^2g)	LMC (%)
Belschmeidia roxburghiana	Lauraceae	Evergreen	Mesophyll	34.42 ± 1.2	107.47 ± 6.11	54.882 ± 1
Callophylum polyanthum	Calophyllaceae	Evergreen	Mesophyll	31.37 ± 0.9	120.34 ± 8.41	60.472 ± 1.8
Diospyros lanceifolia	Ebenaceae	Evergreen	Mesophyll	34.23 ± 0.9	105.7 ± 5.1	60.819 ± 1
Ixora walichii	Rubiaceae	Evergreen	Mesophyll	72.5 ± 2.8	136.81 ± 5.7	64.37 ± 1.2
Lithocarpus xylocarpus	Fagaceae	Evergreen	Mesophyll	43.95 ± 2.4	117.86 ± 5.8	52.303 ± 1.4
Quercus spicata	Fagaceae	Evergreen	Mesophyll	64.78 ± 2.4	124.02 ± 7.2	52.484 ± 1
Symplocos racemosa	Symplocaceae	Evergreen	Mesophyll	84.35 ± 2.7	121.82 ± 9.5	63.99 ± 2
Symplocos theaefolia	Symplocaceae	Evergreen	Mesophyll	24.18 ± 1.03	111.2 ± 7.6	59.332 ± 0.9
Syzigium claviforum	Myrtaceae	Evergreen	Mesophyll	40.06 ± 1.7	101.282 ± 8.9	55.354 ± 1.3
Thing chhunghul		Evergreen	Microphyll	16.23 ± 0.5	184.25 ± 15	62.356 ± 1.5
Schima walichii	Theaceae	Evergreen	Mesophyll	62.47 ± 2.2	154.984 ± 4.8	63.554 ± 2.3
Dipterocarpus retusus	Dipterocarpaceae	Deciduous	Mesophyll	21.5 ± 0.8	107.41 ± 10.3	55.618 ± 1.1
Engelhardtia spicata	Juglandaceae	Deciduous	Mesophyll	50.89 ± 1.4	156.1 ± 12.8	64.349 ± 2
Eriobotrya bengalensis	Rosaceae	Deciduous	Mesophyll	125.05 ± 4.3	93.25 ± 6.1	57.012 ± 1.4
Helicia excelsa	Proteaceae	Deciduous	Mesophyll	45.93 ± 1.5	122.27 ± 7.3	61.946 ± 0.6
Quercus floribunda	Fagaceae	Deciduous	Mesophyll	44.57 ± 2.06	119.415 ± 9.7	48.734 ± 2.9
Rapanea capitellata	Myrsinaceae	Deciduous	Mesophyll	29.63 ± 1.4	110.07 ± 8.7	64.966 ± 0.8
Ostodes paniculata	Euphorbiaceae	Deciduous	Macrophyll	186.86 ± 7	236.27 ± 20.8	70.394 ± 0.9
Styrax polysperma	Styracaceae	Deciduous	Mesophyll	65.65 ± 1.9	130.5 ± 4.4	63.653 ± 1.2
Embelia tsjeriam	Primulaceae	Deciduous	Microphyll	18.67 ± 0.9	111.55 ± 8.3	64.37 ± 2.7

was fewer under dense growth. Earliest leaf fall started by the month of February and continued till the end of winter, it can be said that senescence can be observed mainly during the end of monsoon and winter season.

The average leaf flush duration was 45 days with a deciduousness day of 30 days. Seasonality is less pronounced in sub-tropical areas in comparison to temperate forest or of dry forest (Borchert et al. 2004, Singh and Kushwaha 2006, Kushwaha et al. 2010, Kushwaha et al. 2011a and b), observable leaf exchange and erratic leaf production occur almost throughout the year for most of the trees studied. This study reflected marked variations in the annual rainfall and temperature during the two annual cycles (i.e. May 2016–April 2017 and May 2017–April 2018). There was an increase of 397 mm of total rainfall during the II cycle with an average monthly increase of 33 mm and a marked increase of 1.9^{o}C in mean temperature during the II cycle.

The study concludes that even within a mild climatic variability significant enter specific variations in the tree phenological patterns occurred in sub-tropical wet forest of Mizoram. Therefore, it is suggested that even a small alteration in environmental condition due to change in climate may significantly affect plant phenology and thus site productivity of this region. A long term phenological studies for various species of the region would be prerequisite to predict the early sign of climate change on tree phenology and productivity of the region. Monitoring plant phenological observation are gaining interest world over as to how climatic alterations affect tree phenology. There is dearth of information on this aspect in this region which is experiencing climate change due to human interferences and therefore urgently needs long term phonological studies.

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