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## REPRODUCTIVE PHENOLOGY OF *LAGERSTROEMIA SPECIOSA* (LYTHRACEAE) IN SOUTHERN WESTERN GHATS, INDIA

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**ABSTRACT:** *Lagerstroemia speciosa* known as “natural plant insulin” effective on the treatment of diabetes due to its insulin-like activity, and because of other bioactive components has been widely exploited from its natural habitat and hence shows a decline in population along with low regeneration. Flowering and fruiting phenology in 10 natural populations across Southern Western Ghats, India was observed to understand the functional rhythm, spatial or temporal differences in the populations and also to understand the reproductive capacity, there by any limiting factors. A total of 97 trees were marked for phenological observations at 10 sites. It was observed that flowering was initiated immediately after flushing and the total period of flowering oscillated between 40 – 120 days. The peak period of anthesis was noticed between 0530 – 0630 hr of the day, which was found to be correlated with temperature and Rh. The total seed output per tree (reproductive capacity) following open pollination was  $1,75,545.82 \pm 11,934.17$ , however regeneration potential was very low. *L. speciosa* shows annual flowering frequency and comes under the subclass intermediate flowering.

**Key words:** ANOVA; Deciduous forest; flowering phenology, fruiting phenology; pollen production, seed dispersal.

## INTRODUCTION

The timing, intensity and duration of flowering in plants dictate the success of their reproductive cycle which in turn has impacts on the population of those animals, pollinators and frugivores, relying on the plant resources resulting from this process [1]. Plant phenology has been most studied in the temperate zone, in contrast, tropical phenology has been an imprecise and often confusing discipline because it has been relatively little studied. Phenological observations provide a back ground information on functional rhythm of plant and plant communities [2]. Phenological studies are important in understanding species interrelations and their interaction with the environment. Hence, generating data for tropical tree species is important as they face significant exploitation rates as observed by earlier workers [3, 4]. It is advantageous to observe the phenology of a species at different microclimates to understand its complete phenological behaviour in nature as abiotic factors especially temperature, Rh and precipitation have specific roles in regulating phenological events [5, 6]. Western Ghats, one of the biodiversity hotspots had witnessed only a few earlier attempts to understand the vegetative and reproductive phenology of trees at various levels in the works of Pandurangan [7], Bhat & Murali [8], Bhat [9], Rajkumar [10], Murali & Sukumar [11, 12], Jose [13], Sundarapandian *et al.* [14]. The study has been designed to understand phenological attributes, anthesis, pollen production, fruit setting and seed dispersal. The major objectives of the study were (i) to identify the distinguishing phenological features at different localities with possible means of variation (ii) to relate anthesis to the time of the day and associated weather conditions (iii) to document pollen production variation between individuals of *L. speciosa* at different sites. These studies are of great value for theoretical model building as well as for resource management concept development, which is widely applicable in many fields of plant breeding, silviculture, and plant growth rate assessment.

## MATERIALS AND METHODS

### Study species

*Lagerstroemia speciosa* is native to Tropical Asia and distributed in Cambodia, China, India, Indonesia, Laos, Malaysia, Myanmar, Philippines, Thailand and Vietnam [15, 16, 17]. It is a small to medium sized semi-deciduous tree (7 – 21 m tall) with wide spreading crown. In India the plant is distributed across the Northern Himalayas and Western Ghats.

The natural populations are seen along the Southern Western Ghats predominantly in the semi-evergreen and moist-deciduous forests of Kerala portion. The tree is a potential source of Corosolic acid, which is known for its anti-diabetic and anti-obesity activity [18]. It also possesses compounds like alpha- and beta-pentagalloylglucose ( $\alpha$  and  $\beta$  PGG) with health-beneficial bioactivities such as anticancer [18], anti-HIV [19], anti-SARS [20], anti-inflammation [21, 22] and antioxidant activity [23]. The plant extracts also possess Anti-fibrotic effect [23] and Antinociceptive Activity [24]. The plant is found to be exploited due to its many fold economic importance resulting in the fast decrease of available strands resulting in a decrease of its genetic diversity. In spite of this only a few attempts have been made pertaining to the phenology/reproductive dynamics of this species which is necessary for a successful management plan.

### Study area

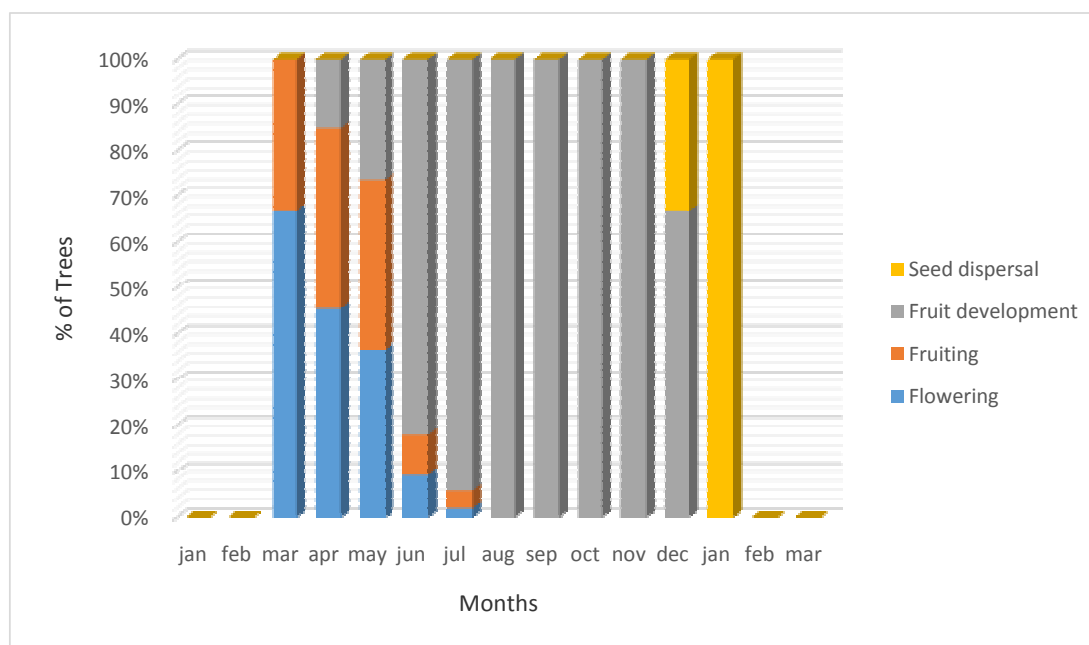
The investigations were carried out at different elevations at ten localities marked as monitoring plots in the Western Ghats as detailed to represent the whole array of variations in temporal differentiation among phenophases due to microclimatic conditions [Sasthanada: 45.72 m a.s.l., 8°53' N, 77°03' E (N = 10); Edapalayam: 46.9 m a.s.l., 8°57' N, 77°08' E (N = 6); Kulathupuzha Pop I: 37.5 m a.s.l., 8°52' N, 77°02' E (N = 10); Kulathupuzha Pop II: 43 m a.s.l., 8°52' N, 77°02' E (N = 10); Perunthenaruvi: 17 m a.s.l., 9°24' N, 76°52' E (N = 10); Chaliarmukku: 30 m a.s.l., 11°18' N, 76°14' E (N = 10); Nellikuthu: 80 m a.s.l., 11°21' N, 76°20' E (N = 9); Thalkolli: 51.5 m a.s.l., 11°16' N, 76°21' E (N = 10); Pattakarimba: 75 m a.s.l., 11°10' N, 76°19' E (N = 10); Mukkali: 526 m a.s.l., 11°03' N, 76°32' E (N = 10)]. There are three main seasons: winter (December – February) which is cold and relatively dry; summer (March – Mid June), warm and dry and rainy (June – September), humid and wet. The mean temperature of the study areas are 22 to 37 °C with an annual mean rainfall of 830 mm.

From January 2011 to December 2013, phenological observations were made at each site for the permanently marked trees, taking into account the occurrence, duration and frequency of the following events: flowering (from floral buds initiation to withering of flowers); fruit set (green fruit to mature fruit to seed dispersal) following Opler *et al.* [25] and Fournier & Charpantier [26]. A particular phenophase was considered to have started when 10% of individuals was observed in that phase and considered to have been completed when less than 10% individuals remained in that particular phase.

The number of inflorescences per tree and number of flowers per inflorescence were counted. Daily rates of anthesis were recorded on mature floral buds of the inflorescences. From each tree five twigs were marked for counting the opened flowers at a time interval of 1 hr over a day. The investigations on the floral biology mainly concentrated on the determination of the number of anthers/flower, number of locules and ovules per flower, pollen ovule ratio, pollen viability, number of stamens and number of flowers opened per inflorescence in a day. Twenty flowers were used to obtain the ovule number per flower, after dissection of the ovary under stereomicroscope. To estimate the number of pollen grains per flower, three anthers of five flowers were separately picked and macerated with a solution of distilled water and detergent. Pollen count was studied following Shivanna & Rangaswamy [27] and Cruden [28]. The effect of time, canopy temperature and RH of the day on the flower opening was examined using ANOVA, with time, temperature and RH as fixed effects – independent variables. The changes in the size of the ovary were observed from the day of anthesis to understand the development of fruit, recording the enlargement on every week.

## RESULTS AND DISCUSSION

Reproductive phenological events of *Lagerstroemia speciosa*, expressed by the percentage of trees in a phenophase among the marked plants were given in Fig. 1. Initiation of sprouting and development of leaf commenced in the month of March and extended up to May during the years of observation. The shoot continued its development generally up to 8 leaf stage after which the terminal bud turns to inflorescence primordia or remains dormant. After the conversion of the apical bud of a branch into Inflorescence primordia, the first flower bud appeared after seven days, which opened on the 11th day. Fifty percent flowering was observed after 19 – 21 days and full bloom after 65 days. Initiation of flowering was found to be continuous with leaf flushing as it starts afterwards. All the process was found to be earlier by 15 days during 2013 when compared to the previous years as the dry season appeared quite early with a marked increase in mean temperature showing a positive correlation between the appearance of dry period and leaf sprouting. The total period of flowering was found to be 90 – 110 days in *L. speciosa*.



**Fig. 1. Reproductive phenological events of *L. speciosa*, expressed by the percentage of trees in a phenophase**

A tree contained  $20 \pm 6.70$  inflorescences which is 15 to 40 cm long and an average of 0.76 ( $n = 53.84 \pm 22.78$ ) flowers opened per day. Colour of flower varied from pink to purple. Flower yield was found to be highly variable parameter which ranged from 1,076 to 6,080 with a coefficient of variation of 42.32 %. Flowers opened with a slit at the top of the bud that widened gradually along the margins as calyx lobes roll back exposing the petals and took 2 – 3 hr for complete blooming. However the time varied according to weather conditions. The anthesis happens slightly before dawn, between 0530 – 0630 hr. A mean temperature of 24 - 27 °C and relative humidity of 71 - 75 % were recorded during anthesis. It was observed that anthesis significantly correlated with time, temperature and RH (Table 1). In *L. speciosa*, gynoecium was found to be receptive during anthesis and hence it is protogyny in nature. Analysis of pollen production per flower, per inflorescence and per tree over the different sites showed no significant variation ( $R^2 = 1$ ,  $p = 0.437$ ) (Table 2). The mean pollen production was  $779.48 \pm 140.92$  per anther,  $1,30,101.65 \pm 21,283$  per flower with a coefficient of variation of 16.36 %. Considerable variation was observed in the number of inflorescences, flowers, anthers and pollen grains per anther etc. from individual to individual and one season to another. Values for various attributes of reproductive biology of *L. speciosa* are given in Table. 3. The main visitors observed during flowering period were honey bees (*Apis indica*, *A. mellifera*, *A. florea*, *A. dorsata* and *A. cerana*), solitary bees like *Amegilla* sp. and carpenter bees (*Xylocopa* sp.). Some spiders like *Oxyopes shwetha*, *Thomisus lobosus* and *Pecutia* sp., ants like *Polyrhachis* sp. and *Technomyrmex* sp. are seen associated with the plant during the period of flowering as pollen robbers.

**Table 1. ANOVA of the effect of time, temperature and relative humidity of the day on anthesis**

Responsible variable	df	MS	F	P-value
<i>In 2011 (<math>R^2 = 1</math>)</i>				
Temp	4	4.008111	3.148281	0.027285
RH	4	3.537556	2.728952	0.046292
Time	4	3.525	2.473684	0.06731
<i>In 2012 (<math>R^2 = 1</math>)</i>				
Temp	4	6.166111	2.713526	0.047209
RH	4	6.915222	3.179088	0.026256
time	4	7.225	2.997037	0.035394
<i>In 2013 (<math>R^2 = 1</math>)</i>				
Temp	4	2.42611	2.67381	0.04659
RH	4	2.822556	3.296201	0.022699
time	4	2.9625	3.172084	0.028659

**Table 2. Pollen production in *L. speciosa* at 10 different sites**

Site	Pollen grains/anther	Pollen grains/flower	Pollen grains/inflorescence
S1	723.32 ± 123	126508.67 ± 1571.94	6811226.79 ± 35808.79
S2	713 ± 142	124703.7 ± 1814.76	6714047.21 ± 41340.23
S3	689 ± 140	120506.1 ± 1789.2	6488048.424 ± 40757.98
S4	733.59 ± 125	128304.891 ± 1597.5	6907935.28 ± 36391.05
S5	811.39 ± 89	141912.111 ± 1137.42	7640548 ± 25910.43
S6	845.41 ± 76	147862.209 ± 971.28	7960901.33 ± 22125.76
S7	836.21 ± 81	146253.129 ± 1035.18	7874268.46 ± 23581.40
S8	799.45 ± 91	139823.805 ± 1162.98	7528113.66 ± 26492.68
S9	821.12 ± 71	143613.888 ± 907.38	7732171.73 ± 20670.12
S10	777 ± 131	135897.3 ± 1674.18	7316710.63 ± 38137.82

\* Mean ± SD

**Table 3. Various attributes of reproductive biology of *L. speciosa***

Inflorescence per tree	20 ± 6.70
Flowers per inflorescence	53.84 ± 22.78
Calyx/corolla lobes	6 (occasionally 4, 5, 7, 8)
Anthers/flower	174.9 ± 12.78
Pollen per anther	779.48 ± 140.92
Pollen per flower	1,30,101.65 ± 21,283
Pollen per inflorescence	99,68,819.06 ± 16,30,773.26
Pollen per tree	26,61,67,468.89 ± 4,35,41,646.17
Carpels	1
Locules in ovary	6 (occasionally 4, 5, 7, 8)
Ovules/ovary	237.8 ± 27.88
Ovules per inflorescence	18232.53 ± 2137.61
Ovules per tree	4,86,808.56 ± 57,074.05
Pollen ovule ratio	605.18 ± 116.85
Fruits per infructescence	43.63 ± 11.62
Seeds per fruit	119 ± 8.09
Seeds per infructescence	6,574.75 ± 446.97
Seeds per tree	1,75,545.82 ± 11,934.17

\* Mean ± SD

Fruit setting started after 3 days of anthesis with visible change in the size of ovary. Retention of fruits after 50 days of pollination was  $51.27 \pm 14.49$ . The average number of fruits per inflorescence after 3 months was found to be  $43.63 \pm 11.62$ . A mature fruit was  $24.99 \pm 2.56$  mm long and  $19.37 \pm 2.53$  mm in diameter, with a covariance value of 5.62. Length and breadth of fruits showed a uniform trend with regard to extend of variation. The L/B ratio of the fruits recorded the least variation (CV = 5.3). It took around 180 - 220 days to obtain full maturity and then to dehisce. The fruits were generally six loculed but 4, 5, 7 and 8 loculed conditions were also observed. The average weight of fruits was  $5.53 \pm 1.11$  gm. The seeds are albuminous with minute embryos, about  $0.71 \pm 0.11 \times 1.41 \pm 0.42$  cm in dimensions, weighing  $6.16 \pm 1.14$  mg; dark brown, smooth, shining. The fruits of *L. speciosa* present a long development period, with an average of 7 months. One fruit set cycle extended from April to December. Seed dispersal occurred mainly in the winter season (December to February). A full mature fruits about to dehisce is greenish brown to brown and by the end of November/early December they started dehiscing. Being a loculicidal capsule with winged seeds, its dispersal is anemophilous. The seeds fallen on the forest floor was found to be taken away by ants (*Diacamma* sp., *Camponotus* sp., and *Cataulacus* sp.) as a secondary dispersal mechanism.

*L. speciosa* shows annual flowering frequency (only one major cycle per year), and comes under the subclass intermediate flowering (1 - 5 months), as per Newstrom *et al.* [1], classification system for plant phenology on flowering patterns. The plant is characterized by low pollen production, the pollen ovule ratio was  $605.18 \pm 116.85:1$  and hence it depends on pollen vector for pollination. Only 36 % the ovules developed into seeds and the total seed output per tree (reproductive capacity) was  $1,75, 545.82 \pm 11, 934.17$ . However only  $35.49 \pm 11.04$  % seeds contained viable embryo (pers. obs.) which hindrances the recruitment rate, which was found to be quite low, apart from other factors like dispersal of seed during the dry period which reduces the probability of seeds falling on a viable environment. Extended studies like seed development and germination studies are required to understand the niceties for low germination rate.



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