

analysis carried out later showed the presence of semi-digested traces of termite-prey.

Intake of small prey, such as termites, can be reasonably linked with the functional adaptations of the frog's head and mouth. The sub-conical head is short and stout, and the pointed snout is calloused at the tip (Figure 1 d), as a protuberance, overhanging the mouth, facilitating the fossorial frog to penetrate the subterranean termite niches while foraging underground. The lower jaw, unlike the more rigid upper jaw, is flap-like and flexible. Bordering with the upper jaw it could form a grooved aperture (Figure 1 d) through which the basally attached fluted tongue can be protruded out to stick or suck up the termite-prey from subsoil fissures, or even from underground termite galleries/tunnels.

The feeding ecology of *N. sahyadrensis* appears to be well integrated with the ecology of soil-termites to the extent that the latter could even become the preferred prey of the frog. Most moist rainforests have relatively large number of genera of termites⁴, and many termite species are subterranean insects colonizing in their abundant population/biomass, thriving on dead wood and humus in the predominantly monsoon type climate of the region. The burrowing and mound-building activities of termites increase the rate of percolation of rainwater and aeration of both the top and subsoil keeping the underground soil temperature low and the moisture content high^{5,6}. This might become indirectly beneficial for the fossorial frog taxon.

The feeding mechanism enabling the underground subsistence of this fossorial frog is apparently evolved in synergic relationship with the ecology of the subterranean termites, which resolves many puzzles on the frog's tackling of the crucial physiological requirements in the underground environment. In India, *N. sahyadrensis* from the southern Western Ghats may be the only known amphibian species that is a fully underground forager. All other burrowing frogs are either open burrow feeders or diurnal burrow dwellers that are open ground feeders in the night.

Other fossorial frogs, which assume a similar foraging strategy are the species of *Rhinophrynus* (Anura: Rhinophrynidae) inhabiting the sub-humid lowland areas from southern USA, Mexico to Costa Rica of North and Central America, and *Hemisus* (Ranidae: Hemisinae) found in tropical and subtropical sub-Saharan Africa⁷. Although both have no phylogenetic relationship with *Nasikabatrachus*, their member species (e.g. *Rhinophrynus dorsalis* and *Hemisus guttatus*) have striking similarity with *N. sahyadrensis* in some of their structural and physiological adaptive features concerned with the feeding mechanism. Both have robust bodies, short limbs, smooth skin and small head. The enlarged, spade-like inner metatarsal tubercles are used to dig into soil rapidly by lateral movements of the feet. They spend most of their life underground, and feed on termites/ants by the unique method of tongue protrusion through a buccal groove. The snout

pointed with calloused tip can penetrate into termitarium or tunnel. They emerge from underground only after heavy rains. *R. dorsalis* exhibits inguinal amplexus, floating on surface of temporary pools⁷.

The morphological and ethological characteristics of the taxon have enabled it to adapt to the present mode of life. In spite of its structural/physiological limitations related to locomotion, feeding, respiration and reproduction, this species considered as a phylogenetic relict one, still shackles itself to its environment, which accounts for its success. While most of the modern amphibians have exhibited amazing evolutionary diversity, this species has taken less progressive changes in the mode of life that is well answered by its phylogenetic senility.

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Vegetative and reproductive phenophases in *Aesculus indica* Colebr. at two different altitudes in Himalayan forests

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Phenological features of *Aesculus indica* Colebr. were studied in Kumaun Central Himalaya in relation to leaf sprouting, anthesis, pollen production, fruit setting, development and retention, and leaf and fruit drop at two different altitudes. Leaf initiation in this species starts in the middle of February and leaf formation occurs in March. Initiation of flowering was observed during the first fortnight of April at both the sites,

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whereas the total period of flowering oscillated between 47 days (on higher elevation, site S₂) and 51 days (on lower elevation, site S₁). The peak period of anthesis was noticed between 1200 and 1400 h of the day. Pollen production per tree varied from 3.90×10^9 to 9.36×10^9 in 1998 and 7.66×10^9 to 9.76×10^9 in the year 1999. Fruit setting was observed in the last week of May with $67.42 \pm 6.39\%$ (site S₁) and $71.87 \pm 6.92\%$ (site S₂) successful fruit setting after 20 days of pollination. Rapid leaf and fruit drop starts from the end of October to early November, after which the tree remains leafless up to mid-February.

Keywords: *Aesculus indica*, Himalayan forests, peak period of anthesis, phenological features.

THE ecological significance of phenological research lies in the fact that it constitutes a dynamic approach for evaluating plant response to the local environment. Phenological observations provide a background for information on functional rhythms of plant and plant communities¹⁻⁷. Phenological adaptations allow utilization of specific resources, which exhibit temporal periodicity and may lead to temporal separation of species⁸. This has been viewed as a mechanism of niche separation and evolution of other new adaptations⁹. Phenological divergence exposes a species to different environmental characteristics, especially those which have relatively wide elevational distribution.

The phenological events of a species are markedly affected by microclimate, viz. north and south-facing slopes, precipitation (seasonal variation in water availability), altitude and topography. Moreover, with increasing altitude, fall in temperature changes the time of occurrence of various phenophases of a species, as phenological events are frequently controlled by temperature. Each phenophase is scheduled to occur at a certain temperature range, above and below which it is replaced by other phenophases. In dry areas, however, precipitation may be more important for regulating the phenological events than temperature, but even in humid temperate areas, it has some effect. Therefore, it is worthwhile to observe the phenology of a species at different altitudes and microclimates to understand its complete phenological behaviour in nature.

The vegetative and reproductive phenology of tree species¹⁰ and understorey species¹¹ of tropical moist forests of Uttara Kannada district, tropical dry forest of Mudumalai area¹² and mid-elevation wet forest¹³ of the Western Ghats, South India, has been worked out by several workers. Similarly the phenology of trees in subtropical evergreen montane forest¹⁴, shrub species in forest fallows developed after slash and burn¹⁵, and dominant tree species in forest ecosystems at Kang chup hills, Manipur¹⁶, Northeast India was also monitored. The vegetative and reproductive phenology of overstorey and understorey species of Similipal Biosphere Reserve (SBR) located in Orissa, India was revealed by Mishra *et al.*¹⁷. The reproductive phenology of angiosperm plant species in a Sa-

vanna-forest mosaic of Venezuelan Central Plain¹⁸ and in tree assemblages of large Amazonian forest landscape¹⁹ was studied in different forest cover types. The pollination ecology of *Pterocarpus santalinus*²⁰ and pollen release in *Pongamia pinnata*²¹ of Fabaceae in the Eastern Ghats of India have also been worked out. Knowledge of anthesis and pollen production is relevant to the study of pollination, developing a functional model for forecasting pollen concentrations and to understand more about the ecological background of pollen dispersal. Yet attempts for gaining such knowledge are few and the methods employed are not uniform. The available literature on anthesis and pollen production is meagre, except for a few scanty reports²²⁻³⁴.

Aesculus indica Colebr. is a large deciduous tree of temperate climate, which is found growing abundantly on the wide elevational gradients in the hills of Western and Central Himalaya between 1500 and 3000 m amsl. The tree is commercially important and has higher food value as the fruits are fed to cattle and goats and the embryo ground and mixed with flour is eaten by the hill people. The seeds yield the oil which is used for making soaps. The wood is used to make utensils and pots.

In Central Himalaya very few studies have been made pertaining to the phenology of trees³⁵⁻³⁷. Keeping in view the aforesaid facts, this study has been designed to understand the phenological attributes, anthesis, pollen production, fruit setting and leaf drop in *A. indica* at two different altitudes in the temperate region of Kumaun Himalaya. The major objectives of the study were: (i) to identify the distinguishing phenological features at two different altitudes with possible reasons of variations in phenophases; (ii) to elucidate anthesis in relation to the time of the day and associated weather conditions; and (iii) to document pollen productivity variations between individuals of *A. indica* at lower and higher altitudes. Such studies shall be 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, plant growth rate assessment and predicting evolutionary changes. The competitiveness of native plants within a formation could also be judged best through such means.

Kumaun occupies the central sector of Indian Himalaya and the study area (Nainital) lies between 29°7'–29°38'N lat. and 79°27'–79°48'E long. Two sites were selected in two distinct micro-catchments representing Sher Ka Danda (site S₁, altitude 2050 m amsl) as lower and Snow View (site S₂, altitude 2550 m amsl) as higher limits of the *A. indica* growth within the catchments. The two sites were established so as to represent the whole array of variations in temporal differentiation among the phenophases due to microclimatic conditions and to explore the relationship between different phenophases and elevations.

The basic climatic patterns in the area are determined by the monsoon rhythm. There are three main seasons; winter, which is usually cold and relatively dry (mid-

December to mid-March); summer, which is warm and dry (April to mid-June) and the rainy period, which is warm and wet (mid-June to mid-September). The transitional period between summer and winter, and between winter and summer can be recognized as autumn (October to November) and spring (February to March) respectively. The rainy season accounts for about three-fourth of the annual rainfall. The mean annual rainfall ranged from 2430 mm (site S₁) to 2545 mm (site S₂), whereas mean annual temperature oscillated between 14.5°C (mean monthly range of 9 to 20°C) at site S₁, and 11.5°C (mean monthly range of 6 to 17°C) at site S₂.

The study was carried out on the isolated trees of *A. indica* at and around Nainital in Central Himalaya. At each forest site a permanent plot of 0.1 ha, which usually represented the most typical growth conditions of *A. indica* trees and was free from biotic disturbances, was established. Detailed phenological records on 20 randomly chosen individuals of the species were made on four phenophases, namely (i) leaf sprouting, (ii) flowering and anthesis, (iii) fruit setting (development and retention), and (iv) leaf and fruit drop, during two successive years from March 1998 to March 2000. The phenological records were made every week during high activity period of the summer season and at 3–4 weeks intervals during the rest of the year. Observations on larger population in other similar stands were also made to supplement information from the permanent plots. The stands were ordinated according to Bray and Curtis³⁸ on the basis of relative density of tree species. The relative density values were standardized before computing the per cent similarity. The phenological events were categorized following Opler *et al.*⁵. A particular phenophase was considered to have started when 10% of individuals was observed in that phase, and was considered to have been completed when only less than 10% individuals remained in that particular phase.

The daily rates of anthesis were recorded on mature floral buds of the inflorescences from five different twigs of five representative trees (two from site S₁ and three from site S₂) on five successive days. The twigs were marked for counting the opened flowers at every 2 h over a day. The counts on opened flowers were made within a twig and subsequently on all five representative twigs of each tree, by a scoring and removing method to avoid duplication. Air temperature and relative humidity were recorded using a thermohygrometer.

Pollen production was recorded on six different representative trees (three each from sites S₁ and S₂) in 1998 and on same individuals in 1999. In *A. indica* the inflorescences remained grouped in bunches on the main branches. First, the main branches were counted, and then a sample of five branches were selected randomly and a count was made of all their bunches. The number of inflorescences per bunch and subsequently for 20 bunches was taken into account, and the number of flowers per inflorescence was ascertained from 20 randomly chosen inflorescences.

Pollen grains were counted using five anthers from different flowers. The anthers were obtained from closed flowers, kept in 70% ethanol, that were washed in distilled water, and placed in test tubes. They were taken apart with the aid of a glass rod and the pollen grains were suspended in 1 ml distilled water. From this concentrate, a drop of 10 µl was transferred to a microscopic slide and the number of pollen grains was counted under a microscope. Counting was replicated five times and the number of pollen grains per anther was calculated according to Cruden³⁹. In order to estimate the total pollen production per individual tree, first the total number of anthers per tree was calculated. Multiplying the number of total inflorescences to the average number of flowers per inflorescence and to the fixed number of anthers per flower, the result was finally multiplied by the average number of pollen grains produced per anther. Thus, an estimate of the total pollen production per tree was made.

For recording fruit setting, five trees of *A. indica* with two branches each were tagged on each site and observations were made after 20 and 50 days of pollination. Development of fruits was observed regularly, but the final dimensions were recorded after full ripening of the fruits. The length and diameter of the fruits were calculated using a vernier calliper. The average weight and size of fruits and stone were calculated on the basis of ten representative fruits from each of the five trees.

The effect of time period (year) and population on the number of pollen grains per flower, inflorescence and per tree was analysed by means of split-plot ANOVA with nesting. Years and populations were examined as fixed effects. Counts were log-transformed in order to improve normality of residuals and to reduce heteroscedasticity. Similarly, the effect of time (6, 8, 10, 12, 14, 16, and 18 h), air temperature and RH of the day on the number of opened flowers (log-transformed) was examined using ANOVA, with time, temperature and RH as fixed effects – independent variables. ANOVA was performed using the SPSS package.

On the basis of phenological observations^{5,35,36,40} on *A. indica*, the various specific events in the phenophases of the trees from both the altitudes (Figure 1) can be categorized as following:

Leaf sprouting and development: In *A. indica* leaf initiation was started in the middle of February during both the years, when about 1 cm long leafy buds emerge from the tip of some branches first. The higher temperature immediately following the low temperatures may aid in this process by inducing bud bursting, which ultimately gives red-coloured new leaves in March. Some workers have also explained the change in temperature^{3,41,42} and photoperiod^{1,43} as possible triggers of bud break. Site S₁ (lower altitude) starts sprouting about 5–7 days earlier. The colour of the new leaves remains pinkish red or yellowish till the end of May, after which it turns bright green

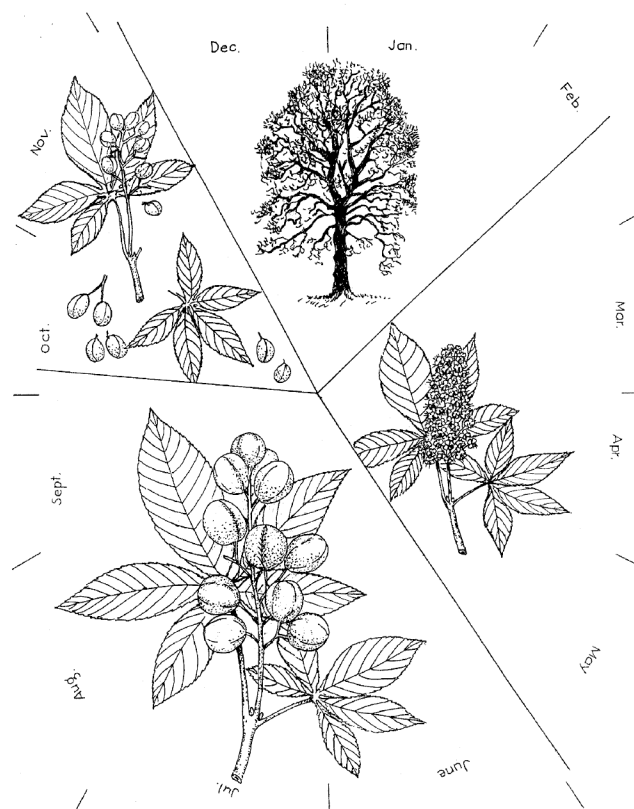


Figure 1. Broad phenophases in *Aesculus indica*.

to dark green up to September. In October, the colour of the leaves turns yellowish-brown to be shed during the deciduous phenophase, which is completed in about 81 to 97 days (Figure 1).

Flowering: The first floral bud in *A. indica* was observed on 7 April at site S_1 and 11 April at site S_2 in 1998. Flowering started from 13 April on the lower site (S_1) and subsequently at the upper altitude site (S_2). Fifty per cent flowering was noticed on 25 April and full bloom on 6 May at site S_1 and almost at a similar time on site S_2 . However, these processes were delayed by one week in 1999 compared to 1998. After this, the flowering activity tapered-off in general with lowest activity at the end of May. The end of flowering at both the sites was on 26–27 May. Thus the total period of flowering in *A. indica* was about 47 days (site S_2) to 51 days (site S_1).

The *A. indica* tree is monoecious in nature. Flowers are white, zygomorphic, horizontal, open in cluster form, in large thyrsoid cyme-bearing terminal panicles. Each flower has crinkled edge. Calyx 0.50 to 0.75 cm long, tubular, with five short, rounded lobes, often split longitudinally in open flower. Petals 4, the place of 5th usually vacant, white and yellow, 1.5 to 2.0 cm long, clawed, unequal in breadth. Stamens 7, filiform, curved upward, longer than the petals; anther versatile. Disk one-sided. Ovary sessile, three-celled; style simple, slender. Nectar cells absent. The anemophilous type of pollination starts in May.

The opening of flowers starts with a slit at the top of the bud that widened gradually; it took 2–5 h for complete opening (in accordance with the associated weather conditions). Maximum anthesis during 1998 and 1999 occurred between 1200 and 1400 h (Table 1), when more than 23 to 43% flowers of the total anthesis within a day had opened at both the sites. Within a single day $11.26 \pm 1.51\%$ to $19.86 \pm 1.74\%$ of total flowers observed on the twig, usually opened. A temperature of 14–30.5°C (in 1998) and 13–31°C (in 1999) and relative humidity ranging from 58 to 62% (in 1998) and 59 to 60.5% (in 1999) were recorded during anthesis. It was also observed that $1.70 \pm 0.89\%$ of the total anthesis was increased during each successive day in both the years (Table 1). Analysis of the number of opened flowers per day revealed highly significant time, temperature and relative humidity effects (Table 2).

The number of inflorescences, flowers, anthers and pollen grains per anther varied considerably from individual to individual and one year to another. Production of pollen grains ranged from 3485.30 ± 89.78 to 3802.00 ± 79.56 and 3342.28 ± 102.13 to 3904.79 ± 98.73 per anther, 24397.10 to 26614.00 and 23395.96 to 27333.53 per flower, and 219573.00 to 239526.00 and 210563.64 to 246001.77 per inflorescence in 1998 and 1999 respectively. Pollen production values per tree varied from 3.90×10^9 to 9.36×10^9 1998 and 7.66×10^9 to 9.76×10^9 in the year 1999 (Table 3). Analysis of pollen production per flower, per inflorescence and per tree over two consecu-

Table 1. Anthesis in *Aesculus indica* on five successive days in relation to time of the day during 1998 and 1999

Date	Buds observed	Percentage of flowers opened at							Total anthesis (%)	Temperature (°C)		
		0600 h	0800 h	1000 h	1200 h	1400 h	1600 h	1800 h		Maxi- mum	Mini- mum	RH (%)
1 May 98	84 ± 5.84	–	7.50 ± 2.95	19.16 ± 2.81	30.83 ± 2.81	31.66 ± 5.83	10.85 ± 3.82	–	11.26 ± 1.51	26.0	15.0	62.0
2 May 98	89 ± 6.72	–	12.50 ± 1.61	17.15 ± 2.39	29.26 ± 1.86	33.82 ± 2.54	7.27 ± 2.41	–	14.23 ± 3.19	25.5	14.5	59.5
3 May 98	78 ± 8.62	–	8.76 ± 1.39	14.64 ± 1.78	31.45 ± 2.13	36.26 ± 2.43	8.87 ± 1.17	–	18.51 ± 1.38	27.5	14.0	60.0
4 May 98	92 ± 10.23	–	9.09 ± 0.94	15.18 ± 1.32	27.27 ± 1.21	34.24 ± 2.14	12.10 ± 1.63	2.12 ± 1.67	19.43 ± 1.59	29.0	14.5	58.5
5 May 98	74 ± 7.12	–	11.62 ± 1.61	18.53 ± 2.10	26.67 ± 1.76	31.95 ± 1.86	8.96 ± 1.43	2.27 ± 2.27	19.86 ± 1.74	30.5	15.5	58.0
Confidence limit		–	9.89 ± 2.05	16.93 ± 1.94	29.10 ± 2.21	33.59 ± 1.92	9.61 ± 1.72	0.87 ± 1.02	16.66 ± 2.34			
4 May 99	72 ± 3.17	–	7.50 ± 3.82	12.26 ± 1.24	27.85 ± 1.49	40.12 ± 1.55	12.26 ± 2.4	–	11.67 ± 1.74	27.0	13.0	60.0
5 May 99	79 ± 8.51	–	9.57 ± 4.82	17.85 ± 1.34	23.45 ± 2.11	28.70 ± 3.97	15.63 ± 1.41	–	16.62 ± 0.66	31.0	14.0	59.0
6 May 99	81 ± 8.97	4.79 ± 2.41	8.58 ± 0.25	7.17 ± 0.50	25.76 ± 0.76	31.31 ± 5.22	11.61 ± 3.28	2.77 ± 2.77	18.26 ± 0.94	31.0	15.0	59.0
7 May 99	73 ± 4.37	2.77 ± 2.77	11.61 ± 2.22	20.66 ± 2.64	23.23 ± 0.15	34.84 ± 2.23	9.65 ± 2.91	–	19.81 ± 2.71	31.0	15.0	59.5
8 May 99	76 ± 6.93	–	8.12 ± 0.98	17.69 ± 3.95	28.80 ± 3.02	32.48 ± 3.94	12.98 ± 1.57	–	16.66 ± 0.53	30.5	14.5	60.5
Confidence limit		1.51 ± 1.59	9.08 ± 2.32	17.13 ± 2.21	25.82 ± 1.79	33.49 ± 3.36	12.41 ± 1.95	0.56 ± 1.09	16.60 ± 1.84			

Table 2. ANOVA of the effect of time, temperature and relative humidity of the day on anthesis in *A. indica*

Response variable and source	df	MS	F	P
In 1998 ($R^2 = 0.562$)				
Time	6	18.05	1.56	0.0043
Temperature	6	24.26	2.14	0.0012
RH	6	31.35	2.89	0.0012
In 1999 ($R^2 = 0.667$)				
Time	6	21.56	1.74	0.0032
Temperature	6	26.24	2.38	0.0024
RH	6	36.20	2.98	0.0032

tive years (1998 and 1999) revealed significant year and population effects along with the significant year \times population interaction (Table 4).

Fruit setting development and retention: Fruit setting in *A. indica* started in the last week of May to the first week of June. In 1998, average fruit setting percentage was 67.42×6.39 after 20 days of pollination on site S_1 , whereas it was 71.87×6.92 on site S_2 after the same period. Retention of fruit after 50 days of pollination was only $61.55 \pm 7.17\%$ on site S_1 and $63.0 \pm 5.16\%$ on site S_2 . Similarly, in 1999 on the lower elevation site (S_1) $73.02 \pm 4.44\%$ fruit setting occurred, which was reduced to $61.08 \pm 5.22\%$ subsequently, after 50 days of pollination. On the other hand, at the higher elevation site (S_2) out of $72.17 \pm 11.05\%$ fruit setting, only $62.44 \pm 9.06\%$ fruit retention was observed after 50 days of pollination (Table 5).

By the end of June the fruits were 1.5 to 2.0 cm long. They matured by the end of July. The fruits were one to three-celled capsules with an average weight of 25.30 g. The average size of the fruits after full ripening was 3.8×3.5 cm. They were long, ovoid and rough from outside. The average weight of the stone was 15.30 g. The seeds were ex-albuminous, about 2.5 cm in diameter, dark brown, smooth, shining; hilum about 1.5 cm in diameter. During August the fruits become brown and by the end of September they started dehiscing to disperse the seeds. Fruit development and maturation during summer and the rainy season indicates the retention of photosynthetic organs throughout this period, which maintained continuous supply of metabolites. This factor along with other favourable conditions, like daylength, temperature, etc., possibly favoured the maturation of fruits during this period. This appeared to be a possible strategy of the species to exploit the maximum favourable conditions available during the rainy season for fruit maturation.

Leaf and fruit drop: In the first fortnight of October the leaves turned yellowish brown, and rapid leaf drop started from the end of October to the first week of November in both the years. Leaf drop was completed by the end of November. In December and January, the tree remained

completely leafless (Figure 1). *A. indica* showed concentrated leaf drop in early winter (October–November). This has resulted in the maximum trees completely leafless, leading to dormancy during severe winters (December to February). Further, in the present study it was observed that the percentage of deciduousness has increased from lower elevation to higher elevation, suggesting increased environmental severity. The activity of leaf drop seems positively related with decreasing atmospheric temperature (September onwards). On the other hand, the fruits become brown in colour during August–September and by the end of September, they start dehiscing. Fruit dropping starts more or less along with the leaves, which is completed by the end of November. The trees of high elevation (S_2) dropped their fruits and leaves nearly 7 to 10 days earlier despite the fact that the fruit maturation at higher elevation site (S_2) was delayed for about 10 days. Due to high food and commercial value of fruits, we observed that the mode of fruit/seed dispersal was therefore brought about by both biotic and abiotic means, although biotic means of dispersal were dominant. However, further investigations are needed before arriving at definite conclusions. The late maturation and lengthy retention of fruits provide better chances for wind dispersal in *A. indica* during early winters, when strong, dry winds blow in this region.

Several deciduous species such as *Populus ciliata*, *Acer oblongum*, *Sapium insigne*, *Alnus nepalensis*, *Fraxinus micrantha* and *Aesculus indica* are referred to as early seral species⁴⁴. This may partly explain the occurrence of these deciduous species in small patches (or gaps) in forests with a close canopy or corresponding to patches of bare sites resulting from frequent landslips and related mass movements specially along the courses of streams⁴⁵.

Seasonality (such as temperature, humidity, rainfall, daylength, soil moisture and wind speed) exposes plants to regular periodic changes in the quality and abundance of resources¹. All these factors are known to play a role alone or in combination in triggering phenological changes. The synchronization of flowering with leaf flushing seems to be related to moisture, temperature and photoperiod^{12,14}. At the onset of summer, melting of snow takes place vigorously due to rise in temperature, which acts as a powerful trigger to terminate the prolonged winter dormancy of both vegetative and floral buds³. The protected dormant buds of the aerial shoots suddenly burst out at the advent of this favourable period around mid-February. Besides temperature, isolated rain showers, which break the long, dry spell, do favour the leafing of plants in this region during March–April. In many tropical deciduous forests, increased leaf fall has been linked with the onset of the dry season and leaf initiation prior to the commencement of the rainy season^{3,43}. Contrasting patterns have been reported for several dry tropical forests, where community-wide flowering peaks were associated with the onset of the rainy season^{5,12}. These have been explained on the basis of changes in temperature^{3,41} and photoperiod^{1,43}.

Table 3. Pollen production in *A. indica* during two successive years

Altitude/ tree no.	1998				1999			
	Pollen grains/ anther	Pollen grains/ flower	Pollen grains/ inflorescence	Pollen grains/ tree	Pollen grains/ anther	Pollen grains/ flower	Pollen grains/ inflorescence	Pollen grains/ tree
S ₁ /1	3540.00 ± 93.16	24780	223020	3.90 × 10 ⁹	3398.73 ± 115.14	23791.11	214119.99	8.57 × 10 ⁹
S ₁ /2	3742.00 ± 81.24	26194	235746	6.79 × 10 ⁹	3904.79 ± 98.73	27333.53	246001.77	8.92 × 10 ⁹
S ₁ /3	3631.51 ± 96.54	25420.57	228785.13	9.27 × 10 ⁹	3683.38 ± 74.19	25783.66	232052.94	9.27 × 10 ⁹
S ₂ /1	3485.30 ± 89.78	24397.1	219573.90	9.36 × 10 ⁹	3342.28 ± 102.13	23395.96	210563.64	7.66 × 10 ⁹
S ₂ /2	3802.00 ± 79.56	26614.00	239526.00	8.38 × 10 ⁹	3883.91 ± 83.19	27187.37	244686.33	9.33 × 10 ⁹
S ₂ /3	3474.36 ± 101.42	24320.52	218884.68	8.82 × 10 ⁹	3742.51 ± 89.75	26197.57	235778.13	9.76 × 10 ⁹
Average	3612.53 ±	25287.70 ±	227589.29 ±	7.75 × 10 ⁹ ±	3659.27 ±	25614.87 ±	267200.47 ±	8.92 × 10 ⁹ ±
	55.84	390.92	3518.25	8.60 × 10 ⁸	97.74	684.21	40115.18	3.00 × 10 ⁸

Table 4. ANOVA of the effect of year and population on pollen grains per flower, per inflorescence and per tree in *A. indica*

Response variable and source	df	MS	F	P
Number of pollen grains per flower ($R^2 = 0.6542$)				
Year	1	38.05	14.64	0.0005
Population	1	22.62	4.74	0.0002
Year \times population	1	7.15	1.56	0.0062
Number of pollen grains per inflorescence ($R^2 = 0.7456$)				
Year	1	51.46	26.64	0.0002
Population	1	26.54	5.68	0.0004
Year \times population	1	9.80	2.58	0.0042
Number of pollen grains per tree ($R^2 = 0.6854$)				
Year	1	81.56	40.24	0.0005
Population	1	38.74	8.38	0.0003
Year \times population	1	14.86	3.98	0.0083

Table 5. Fruit setting percentage in *A. indica* under open-pollinated conditions at two different altitudes in 1998 and 1999

Altitude (site)/tree no.	1998		1999	
	After 20 days of pollination (n = 5)	After 50 days of pollination (n = 5)	After 20 days of pollination (n = 5)	After 50 days of pollination (n = 5)
S ₁ /1	65.46 \pm 2.96	59.35 \pm 3.80	71.74 \pm 5.08	58.02 \pm 4.12
S ₁ /2	69.38 \pm 6.88	63.75 \pm 7.50	74.80 \pm 1.79	64.14 \pm 2.52
Confidence limit	67.4175 \pm 6.3859	61.550 \pm 7.1734	73.0175 \pm 4.4351	61.0775 \pm 5.2188
Coefficient of variation	09.6655	11.8925	06.1980	8.7190
S ₂ /1	65.16 \pm 7.47	58.49 \pm 4.66	69.67 \pm 12.93	60.55 \pm 11.91
S ₂ /2	79.76 \pm 2.49	68.53 \pm 0.79	67.38 \pm 14.76	61.24 \pm 12.56
S ₂ /3	70.67 \pm 4.02	61.99 \pm 5.47	79.44 \pm 3.16	65.54 \pm 2.50
Confidence limit	71.8650 \pm 6.9172	63.0033 \pm 5.1621	72.1667 \pm 11.0495	62.4433 \pm 9.0595
Coefficient of variation	12.0290	10.2395	19.1350	18.1318

At the higher elevation site (S₂), one week late sprouting of leaves and floral buds has been attributed to the micro-climatic variations. As the climatic severity persists for a relatively longer period in this site, this leads to temporal variation in growth activity. Similarly, the 7 to 10 days earlier fruit and leaf drop at higher elevation site (S₂) may be due to decreased temperature and increased environmental severity. Cessation of growth and production during winter may be related to several factors, including decline in temperature, daylength and soil moisture⁴⁶ in a climate of marked seasonal variations. Hopkins^{47,48} reported that, other conditions being equal, south to north progression of spring phenophases in the temperate North America is delayed by four days for each degree of latitude northward, for each 5° of longitude eastward and for about 125 m in elevation. This was a general model and with some improvement, it could be usefully applied in resource management⁴⁹.

The physical properties of the soils at both the sites remained more or less the same, which clearly shows that they do not have a direct bearing on the temporal differentiation of phenophases. Therefore, it is stressed emphatically that the phenological activities in *A. indica* are

largely governed by climatic factors. The rise in temperature during early summers induced growth, whereas the decline in temperature during early winters terminated it. As a result, leaf and fruit drop, and dormancy followed.

Reproductive phenology can be described at community level for flowering, fruit maturation and seed dispersal^{1,5,35} and at population and individual levels^{4,9}. Anthesis is an important criterion for judging the onset of pollen release and subsequent dispersal, which is a prerequisite for plant breeding system. The pattern and quantum of daily anthesis reflect the best time and duration of pollination within a day. In *A. indica* diurnal pattern of anthesis has been recorded; the peak period of anthesis was between 1200 and 1400 h of the day, which ultimately refers to the middle-day pattern of pollen release. As postulated by Ingold⁵⁰, wind-pollinated plants shed their pollen almost entirely during daytime because pollen grains are set free under conditions suitable for effective dispersal, since wet cold weather retards the escape of pollen from the anther sacs while dry weather accelerates it. It is an established fact that the air is more turbulent at higher altitudes compared to lower altitudes. The results of the present study reveal that pollen production was slightly more at higher altitude

(site S₂), i.e. 9.36×10^9 pollen grains per tree in 1998 and 9.76×10^9 pollen grains per tree in 1999, compared to lower altitude (site S₁). This ultimately results in relatively more fruit setting, i.e. 68.53 and 65.54% in 1998 and 1999 respectively, at the higher altitude site after 50 days of pollination (Tables 3 and 5).

A. indica is an anemophilous tree species which is characterized by high pollen production, as the vector of pollination it employs, i.e. the wind, is haphazard and not specific. According to Faegri and van der Pijl⁵¹, the anemophily of any taxa is derived, and an increase in the production of pollen occurs in order to compensate for a reduction in efficiency. The total pollen production per plant is useful to estimate the number of pollen grains that could be in the air during a certain season. It can also be used as an estimate of the production of seeds^{29,52} as the efficiency of anemophilous pollination decreases with a reduction in the concentration of airborne pollen⁵³.

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Introgression of broad-spectrum blast resistance gene(s) into cultivated rice (*Oryza sativa* ssp *indica*) from wild rice *O. rufipogon*

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Rice blast disease caused by *Magnaporthe grisea* (Hebert) Barr, is one of the major diseases of the crop. Host specificity as well as genetic instability of rice blast fungus is the major cause for the breakdown of resistance in many varieties over a period of time.

Here we report the introgression of broad-spectrum blast resistance gene(s) from *Oryza rufipogon* into a cultivated rice variety. An accession (Coll-4) of *O. rufipogon*, highly resistant to blast, was crossed with a susceptible line B 32-Sel-4 and the F₁ was top-crossed with another susceptible line B 29-6. The F₂ population of the three-way cross was screened for blast and salinity tolerance. Twenty selected plants, each with blast resistance and salinity tolerance, were intermated. The process of intermating was repeated in the F₂ progenies derived from the first intermated populations. After two cycles of intermating, pedigree selections in the resulting segregating generations were followed under saline conditions. The 42 introgression lines (F₅ generation) were screened for blast reaction under artificial inoculation. Eight lines were observed to be immune, 20 resistant and six lines moderately resistant to an isolate of blast isolated in Andamans. The remaining eight lines, the two *indica* rice parents and check CO39 were susceptible. Two promising lines with higher yield potential were evaluated in multilocation trials (AICRIP) during 1999, 2000 and 2001 at 17 hotspot locations for blast infection. The culture B 90-15 (IET 15420) showed resistance reaction against 14 isolates and moderate resistance against another two isolates of blast indicating the introgression of a broad spectrum of resistance to blast from *O. rufipogon*.

Keywords: Blast resistance gene(s), introgression, *Oryza rufipogon*, *O. sativa*.

BLAST disease of rice caused by the fungus *Magnaporthe grisea* (Hebert) Barr, is one of the most destructive diseases of the crop causing severe yield loss. The fungus can attack the rice plant at any growth stage and can cause severe leaf necrosis and impede grain filling, resulting in decreased grain number and weight. When the last node is attacked, it causes partial to complete sterility. Host plant resistance is the most promising method to minimize yield loss due to blast disease¹. Several major genes with complete resistance to a specific subset of isolate have been deployed in the development of varieties. The resistance in several varieties having major genes for blast often breaks down due to the evolution of virulent races of pathogen after few years of the release of resistant varieties².

Identification of blast resistance genes indicated 40 different loci in rice^{3–5}, including the *Pi9* gene introgressed from *Oryza minuta*. A number of these genes have been mapped on the molecular linkage map of rice^{5–9}. In addition to genes that confer complete resistance through hypersensitive reaction, genes for partial resistance to blast have also been characterized¹⁰. This quantitative resistance is also referred to as field resistance¹¹, slow blasting¹² and dilatory resistance¹³. Partial resistance is characterized by lesions that are typically spindle-shaped but may be fewer in number, reduced in size, slower to develop, or shorter lived. The net effect is a reduced inoculum potential and a

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