

genetically superior genotypes and their correlation with the environmental attributes.

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Explosive pollen release, wind-pollination and mixed mating in the tropical tree *Shorea robusta* Gaertn. f. (Dipterocarpaceae)

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***Shorea robusta* in tropical India is wind-pollinated. Its gregarious occurrence; flowering during the dry season before the leaves unfold; conspicuous drooping inflorescence with pendulous flowers; massivity of blooming; copious pollen production (c. 60,000 per flower); explosive pollen release triggered by moderately gusty winds; pollen entrapment and transport in moderately turbulent wind; long period of pollen viability (c. 50 h); pollen grain size falling within the aerodynamic size range of 20–60 µm; pollen getting dispersed in single units; high pollen concentration in the air; strong protogyny with prolonged stigma receptivity (c. 50 h) – all constitute the adaptations for effective wind-pollination. The flowers are nearly similar in compatibility to auto, geitono and xeno pollen. The style emerges out a day before the unfolding of the corolla, an adaptive device for pollen reception from xeno and geitono sources; presumed cryptic self-incompatibility facilitates outcrossing at this stage of the flower. The mixed mating system and the stigma receptivity extending into day-2 of flower life when the corolla unfolds allow back-up auto-pollination should outcrossing fail.**

SHOREA Roxb. (Dipterocarpaceae) is a tropical genus widely distributed in India, Sri Lanka, Burma and other South-East Asian countries. It is highly valued for its strong and durable wood, resins, oils, camphor and turpentine. Indian forests harbour four species: *S. assamica* Dyer, *S. robusta* Gaertn. f., *S. roxburghii* (= *S. talura*) G. Don and *S. tumbuggaia* Roxb. Of these, *S. robusta* commonly called the sal tree, is the most important. It is the main source of construction and domestic timber and also dammars of India. It occurs extensively in parts of north, east and central India. The sal forests extend from the Himalayan foothills through central India, Madhya Pradesh, West Bengal, Orissa up to Visakhapatnam in Andhra Pradesh, often forming pure stands over extensive areas, and yielding large fruit crop adapted for wind dissemination. In the aseasonal rainforest of Malaysia, *Shorea* is dominant and its species are pollinated by thrips¹, and in the Sri Lankan forests, honey bees, *Apis dorsata* and *A. cerana indica*, are the pollen vectors². In India, *S. robusta* is presumed to be either entomophilous or anemophilous³, and the plant is a high pollen produ-

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cer^{3,4}. During the studies on the breeding and pollination systems of tree species in the Eastern Ghats, *S. robusta* was found not visited by any anthophilous insect except for the exploratory visits of honey bees. In the light of contrasting reports on the type of pollen vector, the floral mechanism was examined in detail and the operation of wind-pollination was found in this tropical tree species. This contrasts with the prevailing notion that wind-pollination is absent or rare in the tropical trees⁵⁻⁷, and thus appears to be of ecological and evolutionary significance.

The study was carried out during 1995–1996. Blooming phenological and floral data including mating system(s), pollen viability and stigma receptivity of *S. robusta* were collected following the methods of Dafni⁸, and Kearns and Inouye⁹. Pollen production was determined following Subba Reddi and Reddi¹⁰, and pollen concentrations in the air were measured at 5 m height using battery-operated rotorod samplers, according to Subba Reddi and Reddi¹¹. Air samples were collected for 20 min between 0945 and 1015 h for five days during the peak blooming period.

Blooming phenology was studied selecting at random 30 individual trees distributed in an area of about 6 ha in the dry deciduous forest at Gummalakshampuram (18°49'N–83°29'E) in the Palakonda Hill range (18°36'N–83°48'E) of the Eastern Ghats. It was observed that the species blooms once ever year, thus having a flowering frequency of one¹². Blooming commences during late February while the trees are bare of leaves, and progresses through March and terminates in early April, with little asynchrony among individual trees. Blooming is massive, spreading over 33–45 days (av. 35 days), with a peak of 7–13 days (av. 10 days). Inflorescence is a racemose panicle with unilateral branching, conspicuous, 10–22 cm long and drooping, and bears an average of 39 ($R = 12$ –80) flowers which mature and open over a period of 10 days ($R = 6$ –13). Flowers are subsessile, pendulous, hermaphroditic, dichogamous with strong protogyny, 2.5 cm across and lack nectar. Flower life counted from the time of style emergence lasts for three days. Sepals are connate at the base, persist and elongate after fertilization of flowers to form wings thus facilitating wind dispersal of the samara fruit. The corolla is an inverted bowl structure. Petals are pale yellow with mild fragrance, but not showy. Each petal tapers and the tapering is recurved and faces upwards. Stamens are 35 in number ($R = 31$ –40), arranged compactly in three whorls; the filaments are 2 mm long. Anthers are linear, 1 mm long and pilose. The number of pollen grains per anther varies from 1450 to 1860, the average being 1700 grains, and per flower it is 59,500 grains. Pollen grains are viable for over 50 h as indicated by fruit-set upon pollination of virgin stigmas with pollen grains stored indoors. Fruit-set obtained with fresh pollen is 100%, 24-h-old pollen 68%, 48-h-old pollen 48% and 66-h-old pollen 8%. Similar length of pollen viability is also indicated by the germination index of pollen in 30% sucrose solution. Ovary is superior, syncarpous, with three

locules each containing two ovules. Style is relatively long, bearing slightly tridenticulate stigma. It projects out about a day before the petals expand, and the stigma is receptive from then onwards for 50 h as indicated by its viscosity and glistening. Almost the same length of receptivity period is indicated by the fruit-set capability of stigmas of different ages in emasculated and protected flowers. Fruit-set is 100% with fresh stigmas, and it decreased as the stigmas aged. It is 76, 60, 52 and 28% after an interval of every 12 h from the emergence of the style.

Mature flower buds open up at any time of the day, but with a higher frequency at 0800–1000 h and again at 1400–1600 h. About 68% of open flowers are available during light hours and the remaining 32% during dark hours of the day. After making a preliminary observation of anther dehiscence and subsequent pollen release under natural conditions, details were worked out by getting few flowering branches to the forest rest house and maintaining them by placing their cut ends in a bucket containing water and keeping the system in a room with windows and doors kept closed, so that almost still-air conditions prevailed in the room. The following morning from 0730 h, the mature flower buds were observed continuously for recording their opening and anther dehiscence and subsequent pollen release. Microscope slides with their upper surface smeared with white vaseline were also arranged just beneath the expanding flower buds to record the pattern of pollen release. It was observed that dehiscence occurs soon after the opening of the flowers, but there is no pollen release as revealed by scanning the slide under a microscope. Then the flowers were subjected to mechanical disturbance by applying a moderately forceful touch of the finger. Upon mechanical shaking, there was sudden release of pollen, and the pollen deposition on the slide beneath was dense enough to be seen with unaided eye. Observing the slides under the light microscope confirmed pollen release, and most pollen grains got deposited in single units. To know whether moving air could create a similar effect, airflow was generated by operating a table fan for a short period, and the result was affirmative. These experimental observations suggest that the pollen fall is arrested by an inbuilt mechanism until the inflorescence/flowers are shaken by the moving air. Such mechanisms for arresting pollen release until some force shakes the inflorescences are common in wind-pollinators^{13,14}. Probably, in *S. robusta* pollen grains may be held by cohesive and adhesive forces and also the mechanism may be under the control of anther appendage. When the flowers are shaken by the wind force, the tensions are released resulting in explosive release of pollen; and the stigma receives a heavy load of its own pollen.

This pollen release mechanism prevents wastage of pollen by simple gravitational fall from the pendulous flowers and ensures pollen entrapment in moving air and further transport, with no lag between the two events,

thus satisfying an important condition for the efficiency of wind-pollination¹⁵. Pollen concentrations in the air attained 300–1800 per m³ of air at 1000 h. The times of greater proportion of flower opening (0800–1000 h; 1400–1600 h) normally experience moderate turbulent atmospheric conditions, which favour efficient transport of the entrained pollen¹⁶. The transport distance may be long or short, but most experimental evidence suggests a rapid attenuation of pollen concentration, 99% of it getting deposited within a kilometre of the source¹⁷. Accordingly, wind-pollination is generally treated as a short-transport-distance phenomenon and the time gap between pollen release and reception is anticipated to be short; such assumption is also based on the short viable period of pollen grains^{18,19}. However, source height influences transport distance, the distance increasing with the height of the source^{20,21}. As such, relatively long-distance transport is likely to occur with the pollen of *S. robusta*, a large tree of height 18–30 m. Pollen longevity extending over 50 h favours this possibility. Similar longevity of pollen also occurs in other wind-pollinated tropical tree species like *Emblica officinalis* and *Cicca distichus*²². Thus, explosive release of pollen, prolonged pollen viability and tallness of trees may facilitate a relatively long-distance pollen transport. Pollen grains conforming to the typical aerodynamic particle size of 20–60 µm, also permit effective wind transport and favour their deposition on the stigma through impaction^{20,23}. Additional adaptations that contribute to the efficiency of wind-pollination in this species include flowering before full leafing, gregarious occurrence permitting a high density of conspecifics, high pollen production, and more open flowers available during the daytime. Pollen grain exine being reticulate and some of the muri being discontinuous and giving canalculated appearance may contribute to reduced terminal velocity, thus aiding increased dispersal range of pollen¹⁵.

Breeding experiments revealed a mixed system with nearly similar and high auto- (95%) and geitonogamous (91%) and xenogamy (90%). Apomixis is absent. Protogyny of the flowers is a character associated with self-compatible wind-pollinators, and may be a mechanism to reduce selfing²⁴. Recently, more thought has been given to the dilemma of geitonogamous selfing in plant species possessing mixed, flexible or balanced mating system²⁵, and the frequency of such selfing increases with plant size²⁶. Since *S. robusta* is a large tree with massive flowering, there may be a greater frequency of geitonogamous self-pollinations. However, it is argued that long-lived species like the sal tree should strongly favour outcrossing^{27,28}. Then the mechanism favouring outcrossing in the sal tree with mixed mating system must be cryptic in the sense of Bateman²⁹. Probably, a weak self-incompatibility, the presence of which is not revealed in the usual testing procedure may be present and this could only be functional when the xeno-pollen competes with geitonogamous pollen on

the receptive stigma. Evidence is accumulating as to the capability of plants to distinguish between pollen grains of different origin and genetic relatedness³⁰. This form of cryptic self-incompatibility may be ubiquitous in angiosperms³¹. The same may be the single physiological mechanism promoting xenogamy and the associated genetic variability in the sal tree.

Though protogyny does not serve to prevent geitonogamous selfing, it is altogether not irrelevant in *S. robusta*. A heavy load of auto pollen gets deposited on the receptive stigma in the process of explosive pollen release. But for strong protogyny expressed well before the unfolding of the petals, there may not be any space for xeno-pollen on the stigma. Such avoidance of stigma-clogging is also a function attributed to protogyny²⁴. Style projection before the unfolding of the corolla is an excellent device to protect the stigma from overloading with auto-pollen deposition thus permitting the deposition of geitonogamous and/or xeno-pollen, which may result in outcrossing through cryptic self-incompatibility as discussed above. Further, when cross-pollination fails, back-up auto-pollination is facilitated because of prolonged stigma receptivity entering the second day of flower life when the corolla unfolds and pollen is released.

Honey bees, being catholic in their foraging, may visit the flowers of *S. robusta* for pollen collection only after the unfolding of the petals. The bee activity may cause dislodgement and deposition of pollen on the stigma, which might have received geitonogamous and/or xeno-pollen well before the availability of auto-pollen. Thus, bee activity is of no use to *S. robusta*. Such bee visits without benefit to the anemophiles are common³². For the same reason, the visits of thrips, if any, are of no advantage to *S. robusta*.

Earlier studies of pollination mechanisms in the tropical tree species documented wind-pollination in *Madhuca indica*^{23,33}, *Mimusops elengi*³⁴, *C. distichus* and *E. officinalis*²¹ from south India, and in *Trophis involucrate* in the understory of the lowland tropical rainforest in Costa Rica³⁵; now *S. robusta* joins this short list. Interestingly, *M. indica* and *E. officinalis* occur in the same habitat along with *S. robusta*. These proven and confirmed cases of wind-pollination of the tropical trees suggest that geographic distribution of wind-pollination is not as constrained as suggested earlier. Reduced plant diversity and pollinator depauperate conditions are often cited as the selective factors for wind-pollination in the tropical trees^{6,7,36}. Though *S. robusta* might have affected vegetation diversity because of its gregarious character, the study area does not seem to be low in pollinator activity, and anthophilous insects like bees, butterflies, flies and wasps have been found foraging actively on several co-flowering plant species, including trees like *Careya arborea*. Altitudinal factors are also invoked in the adoption of wind-pollination, the frequency increasing with altitude, and

the high Andean genus *Espeletia* responding to altitudinal changes³⁷. To see whether the same factor has compelled *S. robusta* to depend on wind for pollen transport requires complete knowledge of the operating mode of pollination throughout its distributional range. Such a study is worth undertaking in view of the contrasting reports implicating thrips and bees in accomplishing pollination in the species of *Shorea*. Such a study also enables us to understand whether different provenances of this wide-spread species in India have adapted to different pollination modes.

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