



## Reproductive Biology of *Clerodendrum phlomidis* (Lamiaceae)

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### ABSTRACT

*Clerodendrum phlomidis* (Lamiaceae) is an ornamental ever green shrub. Flowering occurs during the period between October - February. Flowers are creamy white or pale yellowish, pedicellate, pentamerous, zygomorphic, hermaphrodite and hypogynous. Anthesis occur between 2030 – 2130hrs followed by anther dehiscence at 2200 – 0130hrs. Stigma becomes receptive at 0300 – 0700 hrs. There are  $3980 \pm 600$  pollens/flower. There are 4 ovules/flower and pollen – ovule ratio is 990: 1. There is no fruit set in bagged inflorescence indicating cross pollinating nature. Fruit and seed set percentage is very low. *Apis cerana indica* and *Apis dorsata* (Honey bee) and butterflies (*Pieris* spp.) are most effective pollinator.

**Keywords :** *Apis cerana indica*, *Apis dorsata*, *Pieris*, protandrous

Reproduction is the only life processes which ensure the perpetuation of life. For successful cultivation and conservation of plants a detailed knowledge of their reproductive biology is required (Moza and Bhatnagar 2007). Reproductive biology of angiosperms has its focus on phenology, pollination biology, pollen-pistil interaction and breeding systems. These are valuable for basic and applied research, having implications to ecological and evolutionary studies as well as agriculture and conservation biology.

The present communication deals with the phenology, floral and pollination biology and breeding system in *Clerodendrum phlomidis* plants growing in different parts of Agra city. The genus *Clerodendrum* was generally placed in *Verbenaceae* but is now correctly included in *Lamiaceae* (Harley *et al.* 2004). The family has a cosmopolitan distribution (Heywood 1978) and the enlarged Lamiaceae contains about 236 genera (Harley *et al.* 2004) and has been stated to contain 6,900 (Heywood 1978) to 7,200 (Harley *et al.* 2004) species, but the World Checklist lists 7,534 (2014). *Clerodendrum* was once a genus of over 400 species (Harley *et al.* 2004) but by 2010, it had been narrowed to about 150 (Yuan *et al.* 2010). *Clerodendrum phlomidis*, commonly known as small arni is an ornamental plant usually grown as a hedge plant. It is cultivated for its ornamental as well as medicinal value. The root is used to make a bitter tonic which is given in convalescence of measles and demulcent in gonorrhoea and decoction of the plant is considered as an alternative it have to cure stomach trouble and swelling in cattle.

### MATERIALS AND METHODS

Twenty plants each growing at two different localities of Agra city were marked and observations were recorded. Observations were made on different phenoevents. Floral morphology, floral biology, number of pollen grains/flower and number of ovules/ovary were studied by various methods given by Kearns and Inouye (1993). Pollen size measured with an ocular micrometer under light microscope following the procedure of McKone and Webb (1988). Pollen viability was

checked by *in vitro* pollen germination by hanging drop culture method using Brewbaker and Kwack's (1963) medium, FCR (fluorochromatic reaction) test after Heslop-Harrison and Heslop-Harrison (1972) and 1% TTC (Hauser and Morrison 1964).

The mode of pollination was evaluated by bagging the emasculating mature buds and then pollinating them artificially. Different pollinators, their population, types and visitation rates were recorded.

The floral buds were also bagged to understand the mode of pollination. Flowering phenology was studied periodically by counting flowers on marked plants throughout the flowering period.

The morphology of different floral parts was studied by scanning electron microscopy (SEM). For SEM studies, fresh anthers and pistils were fixed in 3% gluteraldehyde. These were dehydrated through aqueous acetone series, dried with  $\text{CO}_2$  in a HCP-2. Hitachi critical point dryer. The samples were coating with gold (20nm) in a SCD 020 sputter coating Unit (Poloson Equipment Ltd., Waeford, England) and observed in a Philips EM 50, SEM at all India Institute of Medical Science, New Delhi.

### RESULTS AND DISCUSSION

**Phenology**—*Clerodendrum phlomidis* is an ever green shrub of 4-4.5 m in height, stem ashy-grey, and branches pubescent. Leaf fall and leaf renewal takes place simultaneously. Maximum leaf fall occurs during the months of April-June. Leaf renewal is highest during the months of August which continues till February. Flowering takes place during the months of October to February with maximum floral density in December. Fruit formation starts in November-April.

**Floral biology**—The Flowers (average size  $3.34 \pm 0.28$  cm) are creamy white or pale yellowish arranged in racemes or panicle (Figs. 1a, b) and show herkogamous condition (Fig. 1 c). Flowers are small, complete, pedicellate, pentamerous, zygomorphic, hermaphrodite and hypogynous. Calyx 5, gamosepalous, campanulate, glabrous, pale or some

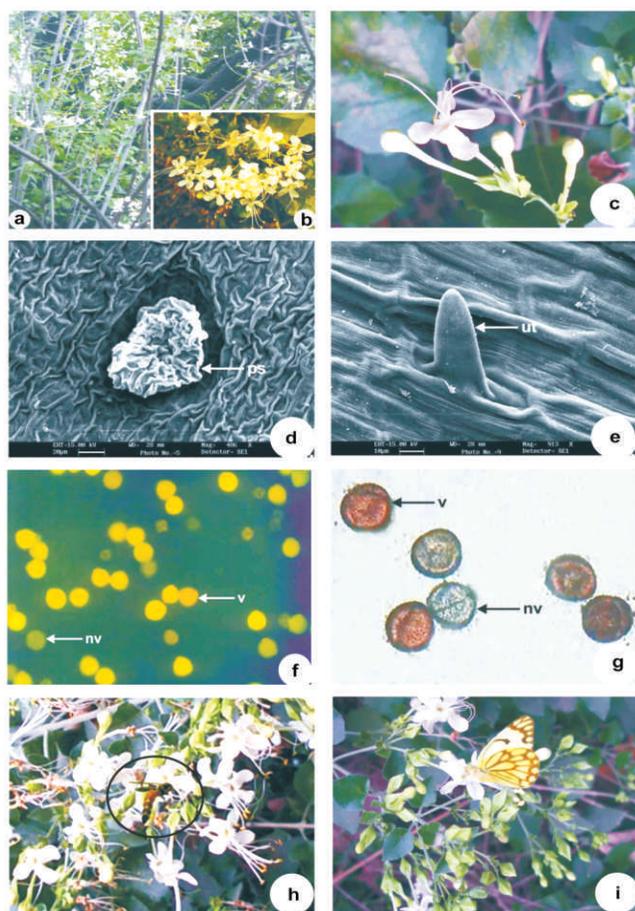


Fig 1.—a. *Clerodendrum phlomidis* plant and inflorescence b. Racemes inflorescence, c. Flower showing bending of stamens and straight pistil, d. SEM micrographs showing peltate scale (Ps) on the surface of corolla (20 $\mu$ m), e. SEM micrographs showing unicellular trichome (ut) on surface on corolla, f. Showing pollen viability as tested by FCR, viable (v), non-viable (nv), g. Pollen viability checked by 1% TTC, viable (v) and non-viable (nv), h. *Apis dorsata* on flower, i. *Pieris* species on flower

what yellowish green. The corolla is much narrower than the calyx, pubescent externally, sub-equal, ovate-elliptic. There are 5 petals, gamopetalous and white in colour. Peltate scale and unicellular trichome are present on surface of corolla (Fig. 1 d, e). Similar observations were observed by Sharma and Rana (2009) in *Duranta repens* another member of the family Lamiaceae. There are four stamens (2.73 $\pm$ 0.15 cm) epipetalous and white in colour. The anthers (0.16  $\pm$  0.04 cm) dehisce through longitudinal slits. Pollen grains are tricolporate, aperture are ectocolpus, long narrow with acute ends (Fig. 1h). Gynoecium is bi-carpillary, syncarpous; ovary bilocular superior, single terminal style; attenuate from the ovary, stigmas bi-lobed; wet type and papillate; Placentation axile, with 2 ovules, one in each locule; ovules are anatropous; unitegmic; tenuinucellate. It forms two types of fruits i.e., true fruit and parthenocarpic fruit. Fruit is an obovoid drupe. Seeds are oblong and white.

**Anthesis**—The flowers are protandrous in nature. They open between 20.30–21.30 h followed by anther dehiscence taking place between 22.00–01.30 h. The stigma becomes receptive between 0300–0700 h. The stigma receptivity was remarkably noticed by hyaline secretion on stigmatic surface. The average number of pollen grain/flower is 3980  $\pm$  600. Pollen-ovule ratio is 990:1.

**Pollen viability**— By FCR test there are 80% viable pollen (Fig. 1f), with 1% TTC test viability is 59% (Fig. 1g) and with Brewbaker and Kwack's (1963) medium it was only 55% with 704 $\pm$ 38  $\mu$ m long pollen tubes. In different sucrose concentration 10% and 20% there was 40% germination with 325 $\pm$ 20  $\mu$ m long pollen tubes and 52% germination with 605 $\pm$ 30  $\mu$ m long tubes respectively.

**Nectar**— At the time of anthesis, each flower produce 3.5 $\pm$ 0.5 ml nectar and the quantity increases and the corolla tube is half filled with 5.5 $\pm$ 0.5 ml nectar at noon and between 1200- 1430 h with 28% $\pm$ 6 sugar concentration. After this period, the quantity of nectar declines. The sugar concentration is not much changed and only in the afternoon there is slight increase in the sugar concentration (30.1% $\pm$ 6). Reddy and Reddi (1995) observed that anther dehisce in the flowers of *Clerodendrum infortunatum* during 0600-0700 h and offer nectar and pollen to insect visitors. Nectar is secreted up to the evening of third day. The nectar sugars are sucrose, glucose and fructose. Sucrose is predominant.

**Pollination biology and breeding system**—Only 8% flowers produce fruits by open pollination. There is no fruit set in bagged flowers indicating its cross pollinating nature and it is self-incompatible. However, in controlled pollination there in 3% fruit formation by geitonogamy and 7% by xenogamy. On the basis of visitation rates, pollen load on their body parts honeybee (*Apis indica* and *Apis dorsata*) (Fig.1h) and butterflies (*Pieris* sp.) (Fig. 1i) are efficient pollinators. The pollen-ovule ratio and hand pollination experiments indicate that it has facultative xenogamous breeding system (Cruden 1977).

Reddy and Reddi (1995) studied butterfly pollination of *Clerodendrum infortunatum*. The breeding system incorporates both geitonogamy and xenogamy. A total of 17 species of insects are found foraging at flowers diurnally. The Papilionoid butterflies (*Papiliopolytes*, *P. polymnestor* and *Atrophaneura hector*) approached the flowers horizontally, grasped it with their legs and continuously fluttered as they probed for nectar. The wings stroke the anthers/stigma, thereby causing pterigotribic pollination. Liza *et al.* (2010) recorded that the *Clerodendrum viscosum* is cross-pollinated and pollinators are black ants, butterflies and long tongue hawk moths. McMullen (2011) made pollination experiments, visitor observations, nectar sampling, pollen transfer studies, pollen-ovule ratio studies, and pollen measurements in

*Clerodendrum molle*, a widespread member of the Galápagos flora. Flowers set fruit and seed via open pollination, autonomous autogamy, facilitated autogamy, facilitated cross-pollination, diurnal pollination, and nocturnal pollination. Cross-pollinated flowers showed a significant increase in seed set over all treatments except facilitated autogamy. Nocturnal and diurnal fruit and seed set did not differ significantly. Nocturnal visitors included ants, spiders, hawk moths, and roaches whereas diurnal visitors included carpenter bees and ants. *C. molle* exhibited incomplete protandry and set fruit via autonomous autogamy as a result of natural selection in an environment with few faithful pollinators. Gautam (2012) studied reproductive biology of *Clerodendrum splendens* an ornamental climbing shrub of the family Lamiaceae. Nearly 2% flower show open pollination. There is no fruit-set in bagged inflorescence indicating cross pollinating nature. *Xylocopa*, *Eumenes* sp. and *Componotus campertris* (Black ant) are most effective visitors.

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