

## **Studies on reproductive biology aspects of *Schizostachyum dullooa* (Gamble) Majumdar (Bambusaceae)**

**C.M. Jijeesh, K.K. Seethalakshmi \* and V.P. Raveendran**

*Kerala Forest Research Institute, Peechi - 680 653, Thrissur, Kerala, India*

**Abstract:** Synchronous flowering was observed in the clumps of *Schizostachyum dullooa* established with rhizomes from Arunachal Pradesh and planted in the Bambusetum of Kerala Forest Research Institute Field Research Centre (N 10° 26' 31" E 76° 21' 36.9") at Veluppadam, Thrissur, Kerala, during August, 2010. Observations were recorded on floral morphology, opening of spikelets, pollination, pollen viability, pollen germination fertilization and post flowering behaviour. Inflorescence of *S. dullooa* is a large panicle and the spikelets were dichogamous, protogynous and open. Abundant pollen production was observed with high viability (96-98 %) and *in vitro* pollen germination. Although, bees visited the flowers and foraged anthers, they did not have any role in pollination. The mode of pollination was anemophily. Flowering was followed by abundant seed and seedling production and death of the culms.

*Keywords:* *Schizostachyum dullooa*, reproductive biology, seed production, pollen viability, post flowering behaviour.

### **INTRODUCTION**

Bamboos, a group of woody perennials belongs to the grass family Poaceae, Subfamily Bambusoideae and Tribe Bambuseae. Bamboo has been a part of human life due to its versatility which makes it a good raw material for numerous applications. A recent compilation reports that there are about 111 genera and 1575 bamboo species in the world. India has the second largest resources of bamboo in terms of species diversity consisting of about 18 genera and 128 species (Seethalakshmi and Kumar, 1998). Bamboo flowering has been amazing to man from time immemorial. Most of the bamboo species are monocarpic and due to the long intermast period for the mass flowering of the majority of bamboo species (Janzen 1976), only a very few reports available on their reproductive biology. *Schizostachyum* is a large genus comprising 45 species distributed in Laos, Malaysia, Philippines, Singapore, Thailand, Africa and India. *Schizostachyum dullooa* (Gamble) Majumdar (dolu bamboo) is a thin-walled sympodial moderate size to large tufted bamboo. It is naturally distributed in

---

\* To whom correspondence should be addressed; E mail: seetha@kfri.org

the moist semi-evergreen forests of northeast India (Assam, Sikkim, Meghalaya, Tripura and Mizoram) to Sylhet, Chittagong and Chittagong hill tracts of Bangladesh. It has multiple uses like, umbrella sticks, flutes, weaving and handicrafts.

In India, flowering of *S. dulloo* has been reported from Singla during 1951, 1957 and 1968 (Gupta, 1972; 1982). Its sporadic flowering is reported from Cachar in 1961 and gregarious flowering in 1971 (Nath, 1962; 1971). Recently, Nath and Das (2010) reported its gregarious flowering in Assam. Synchronous flowering was initiated in 2010 in the *S. dulloo* clumps that were established from rhizomes collected from Arunachal Pradesh and planted in the Bambusetum of Kerala Forest Research Institute at the Field Research Centre, Thrissur, Kerala, India. Other than reports on flowering and floral descriptions, detailed account on reproductive biology is lacking in this species. Hence, investigation was carried out on the reproductive biology (floral morphology, anthesis, pollination and pollen germination) and the post-flowering behaviour.

## MATERIALS AND METHODS

The study was conducted at the Bambusetum (N 10° 26' 31" E 76° 21' 36.9") of Kerala Forest Research Institute, Field Research Centre, Veluppadam, Thrissur, Kerala. *S. dulloo* was introduced in to bambusetum from Arunachal Pradesh during 2004 as rhizomes. The observation on flowering was made in the field during May 2010 to May 2011. Freshly collected inflorescences as well as those fixed in FAA (Formalin Acetic acid Alcohol) were used to record the morphology and dimensions of the floral parts under a dissection microscope (10x). The observations were carried out for six months from August and more number of flowers was observed in field for confirmation. Fifty flowers were randomly tagged and time of anthesis, anther dehiscence and stigma receptivity were observed. Viability of pollen at the time of dehiscence was tested using 1 per cent Acetocarmine, considering stained grains as viable and the shrivelled without stain as non-viable (Radford *et al.*, 1974). *In vitro* germination of pollen was tested in five different germination media (Table 1). Fresh mature anthers were collected from the field at anthesis and pollen grains were carefully dusted in cavity slides containing germination media. Pollen grains were considered to be germinated when the pollen tube length was greater than the diameter of the pollen grain (Tuinstra and Wedel, 2000). The post flowering behaviour of the bamboo species was observed in the field.

**Table 1.** Composition of the pollen germination media

Composition	Media 1	Media 2	Media 3	Media 4	Media 5
Sucrose (g)	10.00	10.00	10.00	0.00	10.00
Boric acid (g)	0.01	0.01	0.00	0.01	0.00
Calcium nitrate (g)	0.03	0.00	0.03	0.03	0.00
Distilled water (ml)	100.00	100.00	100.00	100.00	100.00

## RESULTS AND DISCUSSION

### Flowering phenology and morphology

As an indication of flowering, new culms produced in *S. dullooa* clump during May-June, 2010 were slender and with only half the girth of the culms from the previous year. After the first rains in June, there were 20 old culms and seven new culms in the clump. Signs of flowering were first observed in the culms produced in 2008 (10 culms) in August and two-three culms were in bloom by November. Flowering shoots initiated from the nodes of the branches of flowering culms and elongated up to 12-15 cm, ended with the formation of spikelet at the end and the whole process was completed within four to six weeks. During the early phase of flowering (August to September) the spikelet formation took a longer duration (upto six week as against four weeks during the peak flowering period). The flowers developed in acropetal succession (Fig. 1). Flowering was at its peak during December and January and all the culms were in bloom. During January 2011, fruiting was initiated in the flowered culms. In May-June, the culms stopped flowering and remained in the vegetative phase. Production of new culms also continued in the rainy season and seven newer culms, smaller than the last year culms were produced in June 2011. During August, the culms produced in the previous growing season (2009) started flowering and continued upto December 2011. Flowering of *S. dullooa* occurred between August to December, which in the period in which most bamboo species flower (Banik, 1998, Jijeesh *et al.*, 2009, Seethalakshmi *et al.*, 2010).

The inflorescence of *S. dullooa* is a large panicle. The spikelets were dichogamous, protogynous and open. The occurrence of protogyny was confirmed by the presence



**Figure 1.** A branch of *S. dullooa* bearing inflorescence

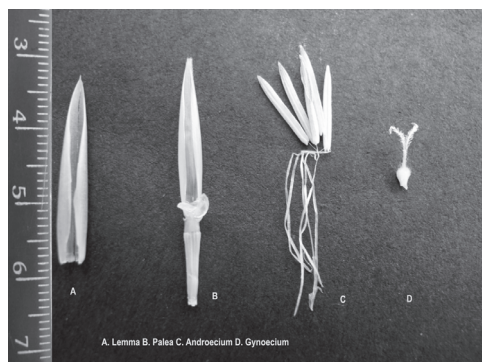
of young anthers along with matured pistil in the FAA fixed flowers as well as maturation of pistil three to five days prior to anther emergence in the field. Dichogamy is observed in most of the bamboo species and protogyny is common (Banik, 1998; Jijeesh *et al.*, 2009; Seethalakshmi *et al.*, 2010). The details of flowering and dimensions of floral parts of *S. dullooa* are given in the Table 2 and Fig. 2. The presence of a steeple-like appendage on the ovary (Fig. 3) was conspicuous.

### Anthesis, pollen viability and pollination

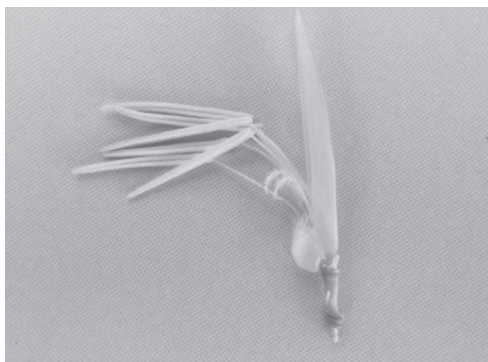
Time of anthesis varied from 6.00 A.M to 12.00 noon. The stigma of the flowers emerged out 3-5 days prior to anther emergence. During anthesis, the stigma receptivity was indicated by the secretion of a viscous fluid and it became receptive by 7.30 A.M.

**Table 2.** Floral characters of *S. dullooa*

Characters	Value
Flower type	Large panicle
Flower colour	Pale green
Length of the panicle (cm)	8.00-12.00
No of florets per panicle	6-8
Number of spikelets with exposed stigma per head	Most of the florets
Number of spikelet with exposed anthers per head	Most of the florets
Number of glumes	4
Length and width of the floret	2.59± 0.45, 0.38 ± 0.09
Length and width of lemma (cm)	2.31± 0.16, 0.36 ± 0.08
Length and width of palea (cm)	1.8± 0.29, 0.42 ± 0.11
Number and length of stamen (cm)	6 , 1.13± 0.05
Anther dehiscence mode	Longitudinal.
Length of the stigma + style (cm)	0.58 ± (0.05)
Stigma type	Three lobed, Wet and papillate
Length of ovary (cm)	0.1
Number of stigma	One
Pollen shape	Round and circular/spherical



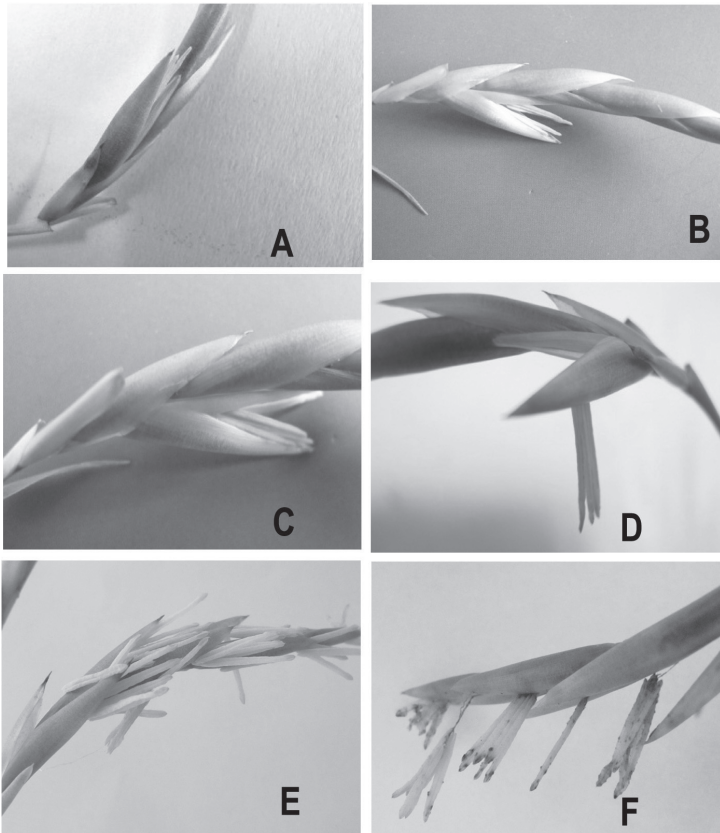
**Figure 2.** Floral morphology of *S. dullooa* showing different parts of a floret



**Figure 3.** The steeple like appendages seen on the *S. dullooa* florets

The yellow anthers emerged out from 8.00 A.M., exerted out completely after 9.30 A.M and linearly dehisced to disperse the yellow monoporate pollens. Gentle breeze shook the anthers to liberate dusty pollen grains in the air. Majority of the anthers curled out in the afternoon (Fig. 4).

The freshly collected pollen grains of *S. dullooa* showed 96 to 98 per cent viability when stained with one per cent acetocarmine. Similarly *in vitro* pollen germination of this species also was found to be high when the pollens grains dusted in different germination media was observed under microscope after one hour of incubation (Table 3). This indicates the production of sufficient amount of viable pollen. The highest pollen germination was observed in the Medium 5 ( $96.8 \pm 1.97$ ) which contained higher sucrose compared to other media. The use of sucrose solution as pollen germination media is generally recommended and the results of the present investigation also agrees with this. The duration of pollen germination was low in the Medium 1 which contained boron and calcium nitrate in addition to sucrose. Lowest



**Figure 4.** The different stages of flower opening in *S. dullooa* from anther peeping out stage to drying

**Table 3.** Pollen germination of *S. dulloo* in different germination media

Media	Germination percentage (%)	Time taken for germination (Min)
M1	72.16 ± 1.8	18
M2	62.8 ± 1.27	26
M3	26.23 ± 2.6	45
M4	6.00 ± 1.9	50
M5	96.8 ± 1.97	28

germination was obtained in the Medium 4 ( $6.00 \pm 1.9$ ) which does not contain sucrose and the duration of germination also was high (50 min). A successful system of *in vitro* pollen germination is a prerequisite for pollen research (Ganeshan *et al.*, 1982) and is important for testing the capacity and viability of pollen for controlled pollinations (Griffin, 1982; Heslop-Harrison, 1979). *In vitro* pollen germination media are needed to study various aspects of pollen biology, pollen selection, pollen transformation and detection of cytoplasmic male sterility system. As many bamboos flower at long intervals and seed production is poor or completely absent in some species, the studies on pollen viability and germination is important. *In vitro* pollen germination rates are considered the best indicator of pollen viability (Shivanna *et al.*, 1991). Abundant seed production in this species can be attributed to the high pollen viability.

Like other bamboo, *S. dulloo* is also anemophilous (wind pollinated). The honey bees, *Apis floria* and *A. dorsata* and some ants were seen visiting the flowers during morning hours. The peak insect visit was observed during 8-11 A.M. after 11.00 A.M. the insect visitation was rare. The presence of large number of pollen grains on the adhesive tapes fixed near the flowers also indicated the anemophily. Moreover, the presence of large anthers producing abundant uniform pollen grains which is the characteristic of wind pollinated species also leads to conclusion of the occurrence of anemophily in this species.

### Post-flowering behaviour

The recurrent visits to the flowering locality confirmed that the flowering of this species is followed by the abundant seed production. The seeds were collected by spreading polythene sheath around the clump, cleaned and 1 kg contained 13700 - 15100 seeds. The seed is a caryopsis, length, width and thickness of the seeds were 23.77 mm, 3.86 mm and 2.99 mm respectively (Fig. 5). A higher rate of seed germination was observed in the nursery beds and seedlings were raised in the nursery. The observation on clump growth during the investigation revealed the following. The culms produced during the growing season of 2008 had an average height of 10-12 m and girth of 12-14 cm, were in full bloom during 2010 and died after flowering and completely dried up. The culms produced during 2009 were in bloom during 2011 (10 culms, height 13 m, girth 12-13 cm). By November 2011, these culms were



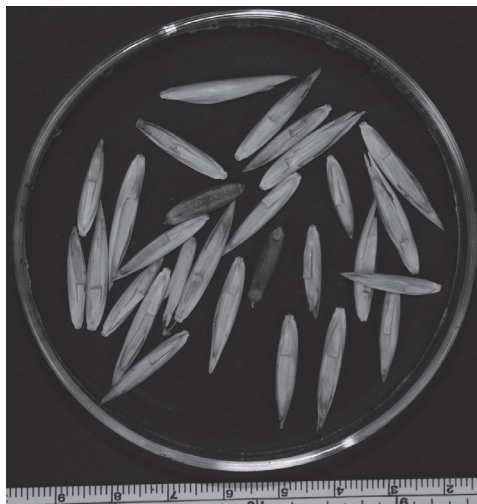


Figure 5. *S. dulloo* seed

leafless, the top portion already dried and flowering was continuing in the bottom portion of the culms. Bottom portion also showed the symptoms of drying. Production of new shoots occurred in the clump during reproductive phase also. The new culms produced during 2010 (May-June) had an average girth of 6-7 cm and 5.2 m height (almost half the size of previous emerged culms). In May- June, 2011, seven new culms were produced (3 m height and 3.8 cm girth). Twenty more thinner culms were produced during September (3 m height and 3 cm girth). It may be concluded that, even though culm production occurs during reproductive phase, the culm size reduced substantially.

Bamboos exhibit different types of post-flowering behaviour like presence or absence of seed production and death of the clumps and flowering and reversion to vegetative phase (Beena *et al.*, 2007; Seethalakshmi *et al.*, 2010). From the present observations it may be concluded that the flowering follows the typical pattern of gregarious flowering where it was followed by abundant seed production and death of the culms.

## CONCLUSION

Unpredictable nature of flowering as well as long flowering cycle of bamboos is still an intriguing concern for researchers. The information on flowering and reproductive biology of most of the bamboo species is limited to occurrence of flowering. As a result of lack of proper documentation on flowering, the age is unknown for many of the major populations of commercial species. This makes difficult of depend on vegetative propagation for large scale production of planting stock. Synchronous flowering of the planting stock with parent plants may result in premature flowering and death of newly established plantation. Since gregarious flowering and seed set is observed in *S. dulloo*, a population of known age can now be raised using seedlings.

## ACKNOWLEDGEMENTS

The authors thank Dr. K. V. Sankaran, Director, and Dr. R. C. Pandalai, Programme Coordinator, Training and Extension Division, Kerala Forest Research Institute for the facilities and National Bamboo Mission, Ministry of Agriculture and Cooperation, Government of India for financial assistance for carrying out the study.

## REFERENCES

- Banik, R.L. 1998. Reproductive biology and flowering populations with diversities in muli bamboo, *Melocanna baccifera* (Roxb.) Kurz, *Bangladesh J. Forest Sci.* 27: 1-15.
- Beena, V.B., Seethalakshmi, K.K. and Raveendran, V.P. 2007. Flowering and reproductive biology of two endemic bamboo species *Dendrocalamus stocksii* and *Pseudoxytenanthera ritcheyi*, *J. Bamboo and Rattan* 6: 11-20.
- Griffin, 1982. A preliminary examination of pollen germination in several Eucalyptus species. *Silvae Genet.* 31: 198-203.
- Gupta, K.K. 1972. Flowering in different species of bamboos in Cachar district of Assam in recent times. *Indian Forester* 98: 83-85.
- Gupta, K.K. 1982. Notes on bamboo flowering in North-East India. *Indian Forester* 108: 596.
- Heslop-Harrison, J. 1979. Aspects of structure, cytochemistry and germination of the pollen of rye. (*Secale cereale* L.). *Ann. Bot.* 44: 1-47.
- Janzen, D. H. 1976. Why bamboos wait so long to flower. *Ann. Rev. Ecol. Syst.* 7: 347 - 391.
- Jijeesh, C.M. and Seethalakshmi, K.K. 2011. Reproductive biology of *Ochlandra wightii* (Munro), an endemic reed bamboo of Western Ghats, India. *Phytomorphology* 61: 17-24.
- Jijeesh, C.M., Seethalakshmi, K.K., Beena, V.B. and Raveendran, V.P. 2009. Recent flowering of an endemic bamboo – *Pseudoxytenanthera monadelpha* (Thw.) Soderstrom and Ellis in Munnar, India. *Phytomorphology* 59: 35-39.
- Nath, A.J. and Das, A.K. 2010. Gregarious flowering of a long lived tropical semelparous bamboo *Schizostachyum dullooa* in Assam. *Curr. Sci.* 99: 154-155.
- Nath, G.M. 1962. Flowering of daloo bamboos in Cachar district. *Indian Forester* 88: 523.
- Nath, G.M. 1972. Flowering of Dalu Bamboo, Assam. *Indian Forester* 97: 498.
- Radford, A.E., Dickson, J.R., Massey and Bell, C.R. 1974 Vascular plant systematics, Harper & Row Publishers New York.
- Seethalakshmi, K.K., Jijeesh, C.M., Beena, V.B. and Raveendran, V.P. 2010. Flowering and post-flowering reversion to vegetative phase of the giant bamboo – *Dendrocalamus giganteus* Wall. ex Munro in Kerala. *J. Non-Timber Forest Prod.* 17 (1): 1-6.
- Seethalakshmi, K.K. and Kumar, M.S. 1998. Bamboos of India: A Compendium, Kerala Forest Research Institute & International Network for Bamboo and Rattan, Peechi, Thrissur, Kerala. 342p.
- Shivanna, K.R., Linkens, H.F. and Cresti, M. 1991. Pollen viability and pollen vigour. *Theor. Appl. Genet.* 81: 38-42.
- Tuinstra, M.R. and Wedel, J. 2000. Estimation of pollen viability in sorghum. *Crop Sci.* 40: 968-970.