Pollen viability, germination and pollination in *Myristica fatua* Houtt. var. *magnifica* (Beddome) Sinclair - a threatened taxon of Western Ghats, India

P RAMA BHAT & K M KAVERIAPPA1

Department of Applied Botany, Mangalore University Mangalagangotri - 574 199, Karnataka, India.

ABSTRACT

A study of anthesis, pollen viability and pollination in *Myristica fatua* var. magnifica showed that anthesis took place early in the morning and anther dehiscence about 36-48 h prior to anthesis. The pollen grains held inside the flower get released as a cloud at the time of anthesis. Male flowers are shed 36-48 h after anthesis. The pollen grains are sticky, pilated in sculpture, spherical and without any visible aperture. Pollen germination commenced 2 h after dusting the grains on slides containing sucrose solution and was completed in about 6 h. Maximum germination (67.3 per cent) was obtained in 5% sucrose solution and the length of pollen tube was also maximum in this concentration. The stained and unstained pollen grains varied in size and the former measured more than the latter. The viability of freshly collected pollen as determined by their stainability in acetocarmine was 94.3 per cent. Among the 10 species of insects collected from trees during flowering, Cremastogaster sp., Sima nigra and Syrphis sp. were most common.

Key words: anthesis, *Myristica fatua* var. *magnifica*, pollen viability and germination, pollination.

Introduction

The family Myristicaceae comprises of 19 genera and about 400 species (Mabberley 1987) of which 7 taxa (excluding the cultivated Myristica fragrans Houtt.) belonging to 4 genera viz., Gymnacranthera, Horsfieldia, Knema - each with a solitary species and Myristica with 4 taxa are reported

from India. The genus *Myristica* includes about 80 species distributed from Tropical Asia to Australia (Mabberley 1987) with New Guinea and the surrounding islands as the centre of distribution (Sinclair 1958). Of the four taxa of *Myristica* occurring in India, *M. fatua* Houtt. var. *magnifica* (Beddome) Sinclair and *M. malabarica* Lam. are endemic to the Western Ghats.

¹Corresponding author

Myristica fatua var. magnifica restricted to Myristica swamp forests, forming its characteristic and dominant species (Krishnamoorthy 1960; Champion & Seth 1968). These swamps are sparsely distributed in Kerala, Tamil Nadu and Karnataka. In Karnataka. they are found only in Gersoppa in Uttara Kannada District. Due to conversion of these swamps into paddy fields and plantations, the population of M. fatua var. magnifica has dwindled to such an extent that it is now classified under the category of rare and threatened plants (Ahmedullah & Nayar 1986). The present paper deals with studies on pollen viability, germination and pollination of this threatened taxon.

Materials and methods

The study was undertaken in *Myristica* swamps located in the evergreen forests at Gersoppa, Uttar Kannada District, Karnataka. Since the plants are dioecious, observations on anthesis were made on male trees, and pollination on female trees.

Ten male trees were selected at random and the time taken from anther dehiscence to anthesis and to shedding of flowers was recorded. The viability of pollen grains was determined by staining them in acetocarmine (Johnson 1940). Pollen grains from mature flower buds and opened flowers were dusted on a clean glass slide containing a drop of glycerine-acetocarmine. The shape, size and surface characters of pollen grains were recroded by sampling about 500 grains collected from 10 trees. About 5000 pollen grains were scored for stainability and the diameter of stained and unstained pollen grains was determined by scoring about 100 grains of each. For studies on germination of pollen grains, six different concentrations of sucrose solution (1%, 5%, 10%, 15%, 20% and 25%) were used. Fresh pollen grains were dusted on a drop of the sucrose solution of each concentration taken on a clean glass slide. The slides were placed in a petri dish lined with moist filter paper and examined up to 22 h, at one hour intervals to determine the time of commencement of pollen germination and completion. The sizes of germinated and ungerminated pollen grains and pollen tube length were measured. About 500 pollen grains were scored for this purpose.

For observations on pollination, flowers of both male and female trees were examined daily from early morning (6 am) till late in the evening (7 pm) from initiation to completion of flowering. The insects visiting the flowers were trapped using a sweep net and preserved in 70% ethanol. Different parts of insects were examined for the presence of pollen grains. The insects were identified with the help of Entomologists of Kerala Forest Research Institute, Peechi; University of Agricultural Sciences, Bangalore and Tamil Nadu Agricultural University, Coimbatore.

Results and discussion

A survey of two swamps in Gersoppa (0.5 ha) showed the presence of 52 adult plants of which 38 were male and 14 were female.

The male plants flowered during November to April and the female during December to March. The mature flower buds opened between 3 am and 5 am. The completely opened flowers were retained on the trees for about 48 h before they were shed. Anther dehiscence took place 36 to 48 h before anthesis and normally dehiscence commenced at

Table 1. Germination of pollen of Myristica fatua var. magnifica in sucrose solution

Concn. of sucrose soln.*	% germination	Mean pollen tube length (μ)
1%	21.56	171.8 ± 15.81
5%	67.30	603.2 ± 24.44
10%	28.81	306.3 ± 16.47
15%	14.15	223.7 ± 17.72
20%	9.39	172.3 ± 10.46
25%	0.00	
Control (Distilled water)	12.25	197.3 ± 8.25

^{*} Incubation was at $26 \pm 2^{\circ}$ C and observations were made 22 h after incubation

9 am and continue up to 2 pm. About 60 per cent of anthers dehisced between 9 am and 11 am. Bavappa & Banda (1981) reported similar observations in nutmeg (*M. fragrans*) where anther dehiscence took place in male flowers about 36 h prior to anthesis.

The pollen grains when observed under the microscope were sticky, spherical, pilated in sculpture and without any visible aperture. The viability of pollen grains as determined by their stainability in glycerine - acetocarmine solution was 94.3 per cent. The diameter of stained pollen grains was slightly more than unstained ones. The average size of the unstained grains was 42,22±3,91 µ, while those of stained ones was 43.72±3.58 µ. The germination percentage was higher in sucrose solution than in distilled water and the highest germination (67.3 per cent) was observed in 5% sucrose. The percentage of germination decreased at higher concentrations. The length of pollen tubes varied in different concentrations of sucrose and maximum length (603.2 ± 16.47μ) was attained in 5% sucrose (Table 1). In *M. fragrans* Bavappa & Banda (1981) reported 96.87 per cent viability by staining with acetocarmine, and 74.3 per cent germination in sucrose solution. They also found differences in the sizes of stained and unstained pollen grains of *M. fragrans*.

Ten species of insects were collected from trees at the time of flowering (Table 2). The most frequently observed insects were Sima nigra, Cremastogaster sp. (Hymenoptera) and Syrphis (Diptera). The legs and proboscis of these insects were found to possess pollen grains. Some of these insects may possibly play a role in pollination of this species. According to Deinum (1949), pollination in nutmeg is carried out by a moth. Bavappa & Banda (1981), however, found no crawling or flying insects visiting nutmeg flowers. They therefore presumed that pollination was assisted by wind. The size of pollen of nutmeg according to them is between media and magna and non-sticky, favouring wind pollination. On the other hand, in M. fatua var. magnifica it is sticky.

Table 2. Insects collected from flowering trees of Myristica fatua var. magnifica

Genus / Species	Family	Order
Cremastogaster dohrni Mayr	Formicidae	Hymenoptera
Cremastogaster sp.	Formicidae	Hymenoptera
Sima nigra Jerdon	Formicidae	Hymenoptera
Moonia albimaculata Dist.	Cicadellidae	Homoptera
Siphonella sp.	Muscidae	Diptera
Syrphis sp.	Syrphidae	Diptera
Triphleps sp.	Anthocoridae	Heteroptera
Unidentified	Chalcidoidea	Hymenoptera
Unidentified	Issidae	Homoptera

Acknowledgements

The authors are grateful to Mr. B V Shetty, Emeritus Scientist, Department of Applied Botany, Mangalore University for critical evaluation of the manuscript. Thanks are also due to Dr. George Mathew, Kerala Forest Research Institute, Peechi; Dr. Viraktamath, University of Agricultural Sciences Bangalore and Dr. Mohan Sundaram, Tamil Nadu Agricultural University, Coimbatore for their help in identification of insects. The financial assistance from Karnataka Power Corporation, Government of Karnataka is also gratefully acknowledged.

References

- Ahmedullah M & Nayar M P 1986 Endemic Plants of the Indian Region Vol. I, Peninsular India. Botanical Survey of India, Calcutta.
- Bavappa K V A & Heen Banda Y M 1981 Anthesis and pollen studies in nutmeg, *Myristica fragrans* Houtt. J. Plantn. Crops 9: 42-45.

- Champion H G & Seth S K 1968 A Revised Survey of the Forest Types of India. Govt. of India Press, New Delhi.
- *Deinum H 1949 Nootmuskaat en foelie - Landb. i, d. Ind. Arch., D 1 2b: 655-683.
- Johanson DA 1940 Plant Microtechnique. Tata McGraw -Hill Publishing Company Ltd., New Delhi
- Krishnamoorthy K 1960 Myristica swamps in the evergreen forests of Travancore. Indian Forester 86: 314-315.
- Mabberley D J 1987 The Plant Book. Cambridge University Press, Cambridge.
- *Sinclair J 1958 A revision of the Malayan Myristicaceae. Singapore Gard. Bull. 16: 205-472.
- * Original not seen