



## Pollen biology and morphology of *Jatropha curcas* L. (Euphorbiaceae)

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### Abstract

*Jatropha curcas*, commonly known as physic nuts and play an important role in biodiesel production and also a food plant of Eri silk worm. The pollen viability, germination and morphology were studied using light and scanning electron microscopy. The pollen viability was found to be  $77 \pm 3.86$  % and maximum germination revealed  $89 \pm 3.24$  % and pollen tube length of  $33.56 \mu\text{m}$ . Pollen grains were monads, spheroidal, inaperture and exine surface Sparsely-reticulate-clavate Crotonoid pattern.

**Keywords:** *Jatropha curcas*, viability, germination, morphology, inaperture, crotonoid, SEM

### 1. Introduction

The genus *Jatropha* belongs to the tribe *Jatrophaeae* of Euphorbiaceae and native of Central America, distributed throughout the world with approximately 175 species [5]. About 11 species *J. curcas*, *J. glandulifera*, *J. gossypifolia*, *J. heynei*, *J. maheswari*, *J. nana*, *J. tanjorensis*, *J. villosa* are available in wild and *J. multifida*, *J. podagrica* and *J. integrriama* are cultivated have been reported in India (www.efloraindia.nic.in).

*J. curcas* is a medium sized tree and distributed in tropical and subtropical forest, which can grow up to 5 meters tall, medicinal and multipurpose plants commonly available in rural areas and also now cultivated in parts in India [13]. It's commonly known as Physic Nut or purging nut, but in different countries and in different language it has different common or vernacular names. The roots, stems, leaves, seeds and fruits of *J. curcas* have been widely used in traditional folk medicine in many parts of West Africa and considered as the most primitive species in the genus [22]. The plant used for resistance to various stresses but also for oil seed crop. The latex of the plant and seed are used in local ayurvedic system of medicine and it's having converting vegetable oil to biodiesel and the fuel properties of the *Jatropha* biodiesel are close to those of fossil diesel and match the American and European standards [21, 10]. In recent years, it has attracted global importance as potential source of biodiesel and breeding programs among different species of *Jatropha* has been undertaken to develop high yielding hybrids [23].

Eri silkworm is a polyphagous sericigenous insect, *Samia Cynthia ricini* feeding on the leaves of a variety of cultivated plants. It exists in the form of nearly 5 ecotypes distributed over different states with varied phenotypic, physiological and behavioral character. *S. C ricini* are found to exhibit preferential feeding habit and luxuriously on the leaves of several food plants including Castor (*Ricinus communis*), Tapioca (*Mamihot esculent*), Papaya (*Carica papaya*) and Barara (*Jatropha curcas*). The host plants of Eri silk worm, in majority are multipurpose plants and have high potential to support various industries in our country [18, 20].

Pollen viability and germination is part of breeding program to develop hybrids and earlier work on pollen biology revealed in the present studies. Li. C *et al.*, [11], Abdelgadir, *et al.*, [1] studied pollen viability and *In-vitro* germination in *J. curcas* and reported pollen from hermaphrodite flowers had a lower germination compare to pollen from male flowers.

Pollen morphological studies in *Jatropha* have been investigated in 11 species, Miller and Webster [14], SEM studies in thirteen species Deghan and Webster [5], apart from few species reported from India by Bahadur, B *et al.*, [3, 4] have described pollen are weakly reticulate pattern without much ornamentation and considered primitive, presence or absence of free processes in lumen and their number varies depending on the species and also clavae/pila are arranged in a regular penta or hexagonal pattern around a depression. Erdtman [6], first studied the pollen of *Jatropha* and proposed the "Crotonoid pattern". Subsequently, many workers reported various *Jatropha* and brought to light the significance of various components of sporoderm ornamentation in relation to taxonomy of the genus. Punt, [15] reported circular not spheroidal and confirm crotonoid pattern suggested Erdtman [6]. Pollen morphology from Brazil Lidian R de Souza, *et al.*, [12], Soto Landeros, *et al.*, [19] from Northwestern Mexico and described the pollen morphology of Euphorbiaceae pollens which are spherical, circular, omniaperturate, densely sculptured with clavate/Pilate processes, aligned articulately to form crotonoid pattern and considered as a unique diagnostic characteristic feature to identify *Jatropha*.

In the present investigation, phenology, pollen viability, germination and pollen morphology were studied in *J. curcas* using Light and Scanning electron microscope.

### 2. Materials and Methods

Field studies were undertaken in different localities of Karnataka to collect Phenological data and recorded in their natural habitats of Ramadevarabetta, Jnana Bharathi campus. The specimens were collected and were authentically identified [16]. For Pollen viability, germination and morphology, fresh material was collected from the wild plants

at Jnana Bharathi campus, Bangalore University, Bangalore.

### 2.1 Pollen Viability and Germination

Flowering twigs were collected from healthy plants during cool hours of the day between 7-8 Am. Fresh pollen was used for germination and viability studies. The pollen viability was tested by staining with 1% 2, 3, 5-Triphenyltetrazolium chloride (TTC) and the percentage of viable pollen were recorded after two hours [17]. The percentage of viable pollen grains was recorded from 5 replications and calculated by the following formula.

$$\% \text{ Pollen Viability} = \frac{\text{No. of stained pollen}}{\text{Total No. of pollen}} \times 100$$

Pollen germination was conducted in different concentration of sucrose ranging from 5, 10, 15 and 20% in Brewbaker and Kawck medium by Hanging drop method [17]. Pollen tube initiation and growth was monitor from 0 hours to 6 hours. The percentage of germination was recorded from 5 replications and estimated by using the formula.

$$\% \text{ Pollen germination} = \frac{\text{No. of germinated pollen}}{\text{Total No. of pollen}} \times 100$$

### 2.2 Pollen morphology (LM and SEM)

Anthers were collected and fixed in 70 % ethyl alcohol and acetolysis was carried out following the method of Erdtman, [7]. Pollen grains were mounted on a clean microscopic slide using glycerin jelly as mounting medium. Measurements of pollen diameters and the structural features of the exine were made with a light microscope.

The acetolysed pollen grains were dehydrated with ethyl alcohol were dusted on the surface of double stick tape pasted on to the stub and coated with gold in a spotter coater. The stub were placed in a vacuum evaporator and observed in the Scanning Electron Microscope (JEOL-JSM840A), Department of Metallurgy, Indian Institute of Science, Bangaluru. The aperture types and surface details were observed.

The pollen general terminology and morphology concepts follow those of Erdtman [6], Hesse *et al.*, [9].

**Table 2:** Pollen morphology of *J. curcas*

Species	Maximum diameter Mean & SD (µm)	Minimum diameter Mean & SD (µm)	Shape	Size class	Colpus type	Exine Sculpturing
<i>J. curcas</i>	60.24±6.04	57.32±5.34	Spheroidal	Large	Inaperture	Sparsely- clavate, Croton pattern

Erdtman [6], described the pollen morphology of *J. curcas* and *J. podagrica* as nonaperturate with croton-pattern exine. Later many Light and Scanning electron microscopic studies have been carried out in different species of *Jatropha* by Punt, [15], Dehgan and Webster [5], EL Ghazaly *et al.*, [8], Bahadur, B *et al.*, [2,3,4], Lidian R de souza, *et al.*, [12], Soto-Landeros, *et al.*, [19] and described the similarity in pollen morphology of the taxa, which are spherical, isopolar, radially symmetrical, circular, omniaperturate, densely sculptured with clavate/pilate head processes, nature of muri, aligned reticulately to form crotonoid pattern but with difference in

## 3. Results and Discussion

### 3.1 Phenology

The plants flowers in two peak seasons in a year, April to May and September to October, but flowering was observed throughout the year. Flowers were unisexual with many male flowers in dichasial cyme and actinomorphic. The male and female flowers ratio varies in different plants.

### 3.2 Pollen Viability and Germination

Pollen was collected at the time of anthesis and viability was found to be 77±3.86 % at anthesis. The pollen germination with various concentrations was observed in the increasing order to that of concentration of sucrose. The minimum pollen germination of 64±2.58 % was observed in 5% sucrose and maximum of 89±3.24% in 15% sucrose. The maximum germinated pollen tube length of 33.56±1.32 µm (table-1). The pollen viability in the present study was comparatively lower (77%) than the previous report of Abdelgadir *et al.*, [1] wherein, they recorded maximum viability of 95.4% in TTC. However, a higher percentage of germination was recorded in the present study of 89 %, while 71.6 % was reported by Li *et al.*, [11] and Abdelgadir *et al.*, [1]. Studies on pollen viability and germination play a crucial role for conservation, interspecific hybridization and management of cultivars.

**Table 1:** Pollen germination, pollen tube length and viability in *J. curcas*

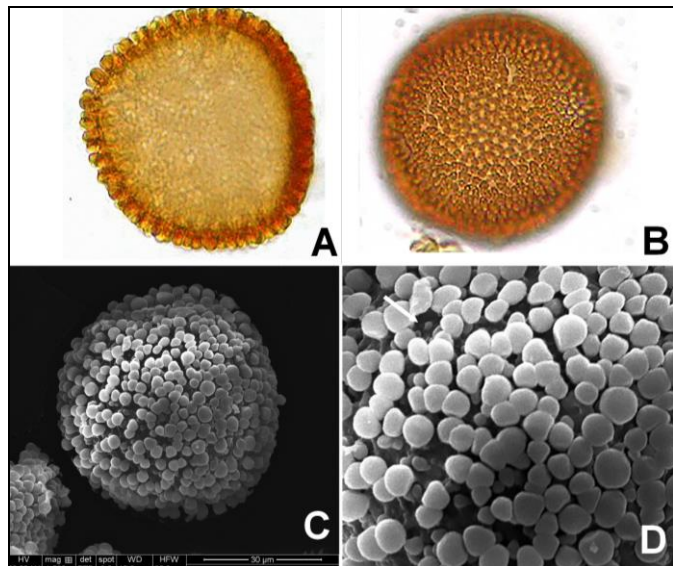
Sucrose with BKM (%)	Germination (%)	Pollen tube length (µm)	Viability (%)
5	64±2.58	33.56±1.32	77±3.86
10	81±2.66		
15	89±3.24		
20	76±2.63		

### 3.3 Pollen Morphology

The pollen grains were monads, large in size with Maximum diameter 60.24±6.04 µm and Minimum diameter 57.32±5.34 µm, spheroidal in shape, exine surface was heavily sculptured, clavate with very few muri irregularly distributed and free standing micro-clavae, inaperturate. Exine pattern is of crotonoid pattern in general (Table-2, Fig-1).

Size and also suggested pollen morphology will contribute to taxonomic and phylogenetic identification.

In the present study all the parameters observed were similar to previous reports except the striations on clavae has observed by Bahadur *et al.*, [4]. Since the pollen grains are inaperturate certain terminologies like polar and equatorial diameter, isopolar and radially symmetrical does not apply as used in previous publications. The observed results differences in the viability and germination percentages and pollen morphology may be due to different climatic conditions of the study sites and ecotypes or genotypes.



**Fig 1:** LM; A- optical section, B- Surface view, SEM; C-surface view D- Enlarged exine surface with muri (arrow).

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