



POLLEN GERMINATION AND TUBE GROWTH IN FOUR SELECTED GENERA OF FAMILY MALVACEAE AND PAPAVERACEAE

Hema Rathore¹, Dushyant K. Singh*², Purti Chaturvedi¹ and Rajneesh K. Agnihotri¹

¹Department of Botany, School of Life Sciences, Dr. B. R. Ambedkar University, Agra, India.

²Department of Botany, Faculty of Science, Bundelkhand University, Jhansi, India

*Corresponding author's email: singh.dks1226@yahoo.com

Abstract

Sexual reproduction in flowering plants comprises pollination, pollen tube elongation, and fertilization. Reproductive biology has been widely applied to the effective conservation and cultivation of species. In the current study, pollen germination and pollen tube elongation were carried out in several chosen plants species of Agra, Uttar Pradesh, for its reproductive success. All plants are helpful the making herbal medicines and belong to family Malvaceae and Papaveraceae. Average pollen germination percentage was determined to be 58% with 55.03 ± 5.01 μm long pollen tubes in *Hibiscus rosasinensis* followed by *Sida acuta* (55%), *Malva maritima* (50 %) and *Argemone maxicana* (47%) with 50.03 ± 4.01 , 55.03 ± 2.5 and 55.03 ± 2.02 μm long pollen tubes, respectively. Pollen grains were found to be three colporate, spheroidal spiny exine, and 4-zonocolpate and reticulate. Under a light microscope, pollen dimension and pollen tube elongation were measured using an ocular micrometre. The present work focused on pollen tube length and pollen germination in chosen plants species in order to its reproductive success.

Keyword: *In-vitro* pollen germination, Malvaceae, Papaveraceae, Pollen tube.

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Introduction

Sexual reproduction in flowering plants is dependent on sperm transport to the egg, which happens via a long polarised projection of a pollen cell known as the pollen tube (Alexander 2007). Pollen germination and pollen tube formation are significant for morphological, physiological, biotechnological, ecological, evolutionary, biochemical, and molecular biology research (Ottavio et al., 1992).

In-vitro germination procedures have been broadly applied on a variety of pollen systems. Such studies have yielded a wealth of knowledge on the physiology and biochemistry of pollen germination and pollen tube formation (Shivanna and Johri, 1985; Heslop-Harrison, 1987). Imbibitions phase, tube initiation phase, lag phase, and fast tube elongation phase are the four phases of pollen germination and pollen tube development (Linskens and Kroh, 1970). Many variables influence pollen tube development, including the temperature, medium osmolarity, and the sucrose, calcium, boron, and zinc are all readily available (Taylor and Hepler, 1997).

Malva maritima, *Sida acuta* and *Hibiscus rosa sinensis* belongs to family Malvaceae growing at different places of Khandari Campus of Dr. B. R. Ambedkar University Agra. These plants occurred in the tropical and sub tropical part of all over India and whole plant has a great medicinal importance which used in the making of herbal medicine. *Hibiscus* is commonly known as China rose or Gurhal. Flower is large, pedicellate, actinomorphic, trumpet-shaped, polypetalous with red velvety long stigma. *S. acuta* as morning mallow or broom weed. Flower is pale yellow in color occurred throughout the year but depending on regional

climate condition. *M. maritima* known as common mallow. Flower is reddish-purple and bright pinkish-purple with dark stripes. *Argemone maxicana* is a herbs species belongs to family Papaveraceae. It is commonly known as Prickly Poppy and Mexican prickly poppy and also known as a pili katheli or satyanashi. It is found in dry soils along roadsides, waste places, open fields and almost every part of India. Flowers occur at the end of branches and yellow in color.

The present work may aid in the provision of important information on the physiology and bio-chemistry of pollen germination and tube elongation which is important for reproductive success.

Materials and Methods

The experiments of an M.Sc. short dissertation submitted in partial fulfilment of the requirement for the degree of Master of Science in Botany were chosen for the present work on pollen germination and tube elongation in four selected genera of the family Malvaceae and Papaveraceae growing at different areas of Khandari Campus, Dr. B. R. Ambedkar University, Agra.

During the flowering stage, pollen grains were collected from newly dehisced anther and tested. Using the hanging drop culture method, the cavity slides are made by suspending fresh pollen grains in a drop of Brewbaker and Kwack's medium (1963). This is the only practical way to track a pollen population over the flowering time (Shivanna and Rangaswami, 1992). The culture medium sterilized by autoclaving. The rim of cavity was smeared with the petroleum jelly. A drop of the B.K. medium was placed on a

cleaned dry cover slip. The drop was suspended in the centre of the coverslip and applied gentle pressure so that the cavity become sealed. The slides were kept in incubator under desired temperature condition. The temperature $22 \pm 2^\circ\text{C}$ is suitable for most pollen species. Under a light microscope, the lengths of pollen tubes were measured with an ocular micrometre (Mckone and Webb, 1988).

Result and Discussion

The present investigation on pollen germination and tube formation has clearly indicated that the Brewbaker and Kwack's medium was the ideal for in vitro pollen germination. The percentage of germination and tube elongation are shown in Table 1.

Table 1 : Pollen germination and tube formation in target plants tested by Brewbaker and Kwack's medium.

Medium	<i>H. rosa sinensis</i>		<i>S. acuta</i>		<i>M. marutiana</i>		<i>A. mexicana</i>	
	PG.	TL (μm)	PG.	TL (μm)	PG.	TL (μm)	PG.	TL (μm)
B.K. Medium	58%	55.03 ± 5.01	55.5%	50.03 ± 4.01	50 %	55.03 ± 2.5	47 %	55.03 ± 2.02

Where, PG-Pollen germination, TL -Tube length

Data show that the average range of pollen germination is 58% with $55.03 \pm 5.01 \mu\text{m}$ long pollen tubes in *H. rosasinensis* when the maximum and minimum temperature range between 12°C - 24°C with 60 % Related humidity. The average size of pollen grain was observed 80-160 μm in diameter, globose to spheroidal. (Figure 1A, 2A and Table 1) Pollen germination is 55% with the $50.03 \pm 4.01 \mu\text{m}$ long pollen tubes in *S. acuta* when temperature range between 12°C - 25°C . The pollen grains were 3 colporate, spheroidal, spiny exine and average size of single pollen grain is 80 μm in diameter, almost half the size of pollen of *H. rosasinensis* (Figure 1B, 2B and Table 1). Pollen germination is 50 % with the $55.03 \pm 2.5 \mu\text{m}$ long pollen tubes in *M. marutina*, when the temperature range between 12°C - 26°C . Average size of pollen grain were observed 50-55 μm in diameter and spheroidal, numerous (Figure 1C, 2C and Table 1). In *A. maxicana* pollen germination were observed 47% with $55.03 \pm 2.02 \mu\text{m}$ long pollen tubes when the temperature range between 18°C - 32°C . The pollen grains were observed 3 zonicolpate, reticulate, monad and znicolpate with rounded ends (Figure 1D, 2D and Table 1).

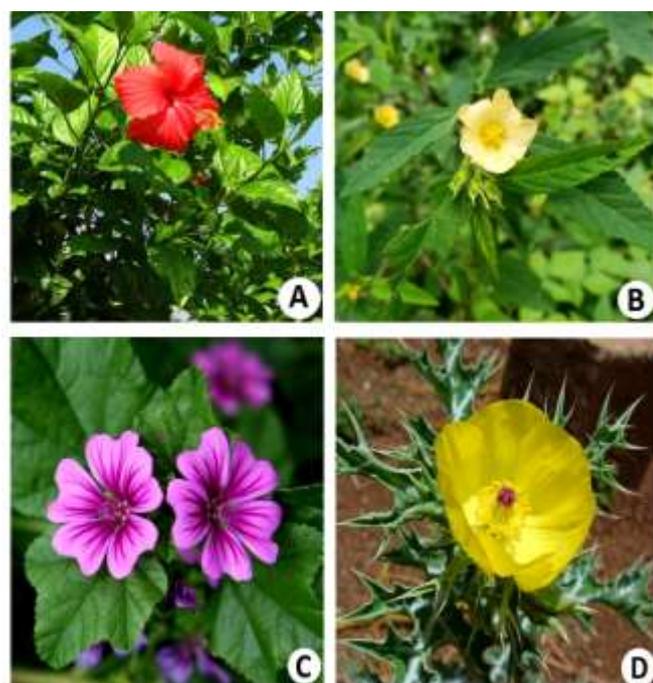


Fig. 1. A: Plant of *H. rosasinensis*, B: *S. acuta*, C: *M. marutiana*, D: *A. mexicana*.

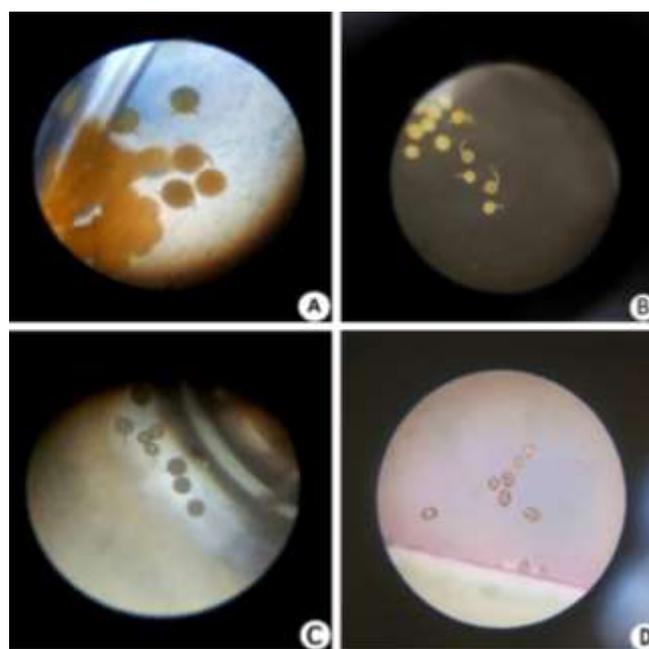


Fig. 2 : Pollen germination and pollen tube length of selected plants A. *H. rosasinensis*, B. *S. acuta*, C. *M. marutiana*, D. *A. mexicana*

To produce optimal responses, the mixture of a germination solution must be empirically formulated for each plant species. Brewbaker and Kwack (1963) medium found to be suitable for most species. Different varieties need different temperature germination. This is following with the results reported in *Trichosanthes dioica* by Kumari *et al.* (2009) and in *Tribulus terrestris* by Ahmad *et al.* (2012). Pollen tube size was more or less determined to be corresponding to the percentage of pollen germination. According to Kumar *et al.* (2010) in *Abutilon indicum* (L) sweet, *in vitro* pollen germination in Brewbaker and Kwack's medium is 56% with $55.03 \pm 6.01 \mu\text{m}$ long pollen tubes. Singh *et al.* (2013) worked in *Sida cordifolia*, he reported that the pollen germination is 56.1% in same medium with $55.03 \pm 6.01 \mu\text{m}$ long pollen tubes. Ahmad *et al.* (2012) working on *Tribulus terrestris* have also reported that increasing the content of sucrose in mixture with 0.01 percent boric acid enhanced the germination percentage.

The present study backs up the findings of Brewbaker and Kwack (1963) by demonstrating that a medium comprising sucrose, calcium nitrate, boric acid, magnesium sulphate, and potassium nitrate is the ideal medium for pollen germination and growth.

Conclusion

In the current investigation, it was observed that the germination and tube elongation is close related to the environment condition such as temperature and relative humidity. Brewbaker and Kwack medium were found to be a suitable medium for germination and development of pollen tube. When boric acid was added to the solution, the percentage of germination and the length of pollen tubes both increased. The present research work could reveal a lot about the biochemistry and physiology of pollen germination and tube elongation, which is important for reproductive success.

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