



EFFECT OF TEMPERATURE AND LIGHT CONDITIONS ON IN-VITRO POLLEN GERMINATION AND POLLEN TUBE GROWTH IN *PONGAMIA GLABRA*, VERT



Vilas A. Patil

Dr. B.N. Purandare Arts, Smt. S. G. Gupta Commerce and Science College, Lonavala, Maharashtra.

ABSTRACT

The present study evaluated the effect of four different temperatures (10, 20, 30 and 40°C) on pollen germination and length of pollen tube growth under in-vitro conditions was investigated in *Pongamia glabra*, Vert. The temperature showed a significant effect on in vitro pollen germination. The highest pollen germination was determined at a temperature of 30°C (63.71%), somewhat lower at 20°C (49.3%) and the lowest at 10°C (31.59 %). Effect of temperature significantly more pronounced on the length of pollen tube. Pollen tube length was higher at the temperatures of 30 and 27°C and was optimal for pollen germination and pollen tube growth. Effect of different light conditions on pollen germination and on length of pollen tube in *Pongamia glabra* was evaluated. Under different light conditions red light showed better effect on pollen germination and on pollen tube growth.

KEYWORDS: BK: Brewbaker and Kwack's medium; TTC: Triphenyl tetrazolium chloride.

INTRODUCTION

Pongamia glabra, Vert. Vern. name Karanj belongs to family Leguminosae sub family papilionaceae is a tree reaching 40-60 ft height; It is used against rheumatism; the oil is also used in soaps. The plant is indigenous to India and cultivated for fibre and for green manure in many states like M.P. Punjab, Maharashtra. The fibre has fair demand from overseas countries and large proportion of it is exported out of India. The crop thrives best in tropical and subtropical climate. The androecium of *Pongamia glabra* are monoadelphous; filaments long and anthers are basifixed. Germination is the first morphogenetic event in the pollen towards fulfilling its ultimate function of discharge of male gamete in the embryo sac. Stigma provides a suitable site for pollen germination. However, studies on in-vivo are not easily feasible because of the complication involve in pistillate tissue. Therefore, the present investigation aimed to study the effect of different temperatures and different light conditions on in-vitro pollen germination in *Pongamia glabra*.

MATERIAL AND METHODS:

The pollen is subjected to viability test with TTC reagent (Triphenyl tetrazolium chloride), I₂KI and acetocarmine stains and the mean value of 10 fields are noted. In vitro pollen germination studies were carried out using mature anther dissected out from the fresh flower of *Pongamia glabra*. In this experiment Brewbaker's and Kwack medium BK medium Boric acid 100 mg l, calcium nitrate 300 mg l, magnesium sulphate acid 200 mg l, potassium nitrate 100 mg l (Brewbaker & Kwack, 1963) was used for culture of pollen. The medium of Brew-baker and Kwack (1963) is one of the most popular for pollen tube culture. Brewbaker and Kwack (1963) tested the pollen of several hundred species, First mount a drop of medium in the cavity of a slide. Dissect fresh pollen from anther and mix in drop of medium. Place the slides in petri

dish on moistened filter paper and cover with lid. Slides were incubated at different temperature and different light conditions. After incubation period drop of cotton blue was added to medium containing pollen grain for germination. Count the number of germinated pollen grains. Note abnormalities and record them, calculate the percent of pollen germination, Length of pollen tube was measured by ocular micrometry and observations were recorded in the observation table. Measurements of pollen tube length were recorded directly by an ocular micrometer fitted to the eyepiece of the microscope. Mean pollen tube length was calculated as the average length of 20 pollen tubes. For each condition ten fields were observed, percentage of pollen germination was calculated by counting germinated pollen grains divided by total number of pollen grains into hundred. This process repeated under different conditions such as light and temperature.

Table No.I. Effect of different temperatures on pollen germination and on length of pollen tube in *Pongamia glabra*.

Germination time: 2 hrs. Relative Humidity: 100%

Sr. No.	Temperature condition	Percentage of pollen germination	Average pollen tube length (μm)
1	Control 27 °C	88.4%	102.21
2	10 ⁰ C	31.59 %	62.51
3	20 ⁰ C	49.3%	74.15
4	30 ⁰ C	63.71%	85.19
5	40 ⁰ C	40.54%	52.82

Table No. II. Effect of different light intensity on pollen germination and on length of pollen tube in *Pongamia glabra*

Germination time: 2 hrs. Relative Humidity: 100%

Sr. No.	Light condition	Percentage of pollen germination	Average pollen tube length (μm)
2	Dark	58.2%	68.8
3	Red	80%	75.6
4	Normal	78.05%	90.9

RESULT AND DISCUSSION:

Pollen viability and fertility was calculated by staining method. Pollen grains from fresh flowers of *Pongamia glabra* which shows just anthesis were stained in I₂KI, Acetocarmine, and T.T.C reagent. The analysis total percentage of viability showed in I₂KI was 94%, in acetocarmine it was 96% while in T.T.C reagent it was 76% at 27⁰c of room temperature by maintaining hundred percent humidity.

The result of pollen viability test given in table number I shows 88.4% germination of pollens of *Pongamia glabra*, Vert. But the germination percentage under different temperature and light condition shows different views (Table I- II). Under different temperature conditions the maximum pollen germination was under 30⁰C. The germination percentage of pollen under 20⁰C was less than those pollen germination under 30⁰C temperature. Secondly the percentage of pollen germination at 10⁰C also less than the 20⁰C and 30⁰C temperature condition. (Table I). It was seen that the average pollen tube length was greater in pollen germination under 30⁰C temperature. The pollen germination under 20⁰C temperature had an average pollen tube length which is more than the pollen tube germination at 10⁰C temperature. Lowest value of pollen tube length was noticed under 50⁰C temperature pollen germination condition. Pollen tube lengths similar to those recorded in the present study were reported for several crops when pollen was grown on artificial media, 1800 μm for corn (Binelli *et al.*, 1985), 450–1400 μm for peanuts (Kakani *et al.*, 2002) Previous studies on pollen germination shown that carbohydrates are responsible for pollen

development and, especially, pollen cytoplasmic carbohydrates and sucrose are involved in protecting pollen viability during exposure and dispersal (Pacini *et al.*, 1996) and for pollen germination, simple sugars are the primary substrates (Stanley, 1971). In pepper plants, exposure to high temperature (32/26 °C) for 8 d resulted in pollen germination of 6 % and shorter pollen tubes compared with maximum pollen germination of 25 % obtained at normal temperature (28/22 °C) (Aloni *et al.*, 2001). In contrast, a decrease in starch and sugar concentration was recorded in tomato pollen grown under high temperature conditions (Pressman *et al.*, 2002). In light condition the maximum germination was obtained under red light. The germination percentage of pollen under dark light was less than those pollen germination under red light. Secondly the percentage of pollen germination under normal light was greater than the germination under dark light but less than red light condition (Table II). It was seen that the average pollen tube length was greater in pollen germination at normal light condition. The pollen germination under red light had an average pollen tube length more than the pollen germination in dark condition. Abnormalities such as bursting; polysiphonism are noticed more or less in all conditions of temperature and different light conditions of light and control conditions. Above results suggest that temperature light plays significant role on pollen germination and growth of pollen tube in *Pongamia glabra*, Vent. under in vitro condition.

REFERENCES:

- Aloni B, Peet M, Pharr M, Karni L.** 2001. The effect of high temperature and high atmospheric CO₂ on carbohydrate changes in bell pepper (*Capsicum annuum*) pollen in relation to its germination. *Physiologia Plantarum* 112: 505–512.
- Brewbaker JL & Kwack BH** 1963 The essential role of calcium ion in pollen germination and pollen tube growth. *American Journal of Botany* 50: 859 - 865.
- Binelli G, De Manincor EV, Ottaviano E.** 1985. Temperature effects on pollen germination and pollen tube growth in maize. *Genetica Agraria* 39: 269–281.
- Kakani VG, Prasad PVV, Craufurd PQ, Wheeler TR.** 2002. Response of *in vitro* pollen germination and pollen tube growth of groundnut (*Arachis hypogaea* L.) genotypes to temperature. *Plant, Cell and Environment* 25: 1651–1661.
- Pacini E.** 1996. Types and meaning of pollen carbohydrate reserves. *Sexual Plant Reproduction* 22: 362–366.
- Pressman E, Peet MM, Pharr DM.** 2002. The effect of heat stress on tomato pollen characteristics is associated with changes in carbohydrate concentration in the developing anthers. *Annals of Botany* 90: 631–636
- Stanley RG.** 1971. Pollen chemistry and tube growth. In: Heslop-Harrison J, ed. *Pollen: development and physiology*. London: Butterworths, 131–155.