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GERMINATION CAPACITY OF STORED POLLEN OF *FICUS CARICA* (MORACEAE) AND THEIR MAINTENANCE

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Abstract

Present investigations pertain to pollen germination and viability of *Ficus carica* L. (Moraceae) for up to 48 weeks. Pollen germination was made by hanging drop technique in different concentration of sucrose and boric acid solutions (10%-100%). The stored conditions are refrigerator, freezer, in vacuum over silica gel and in organic solvents (acetone,benzene, and chloroform). Pollen stored at low temperature (-30°C, -20°C) showed better germination percentage compared to pollen stored at 4°C & fresh. The study indicates that 30% and 40% solutions favoured pollen germination. Benzene showed more germination than acetone and chloroform.

Introduction

Recently pollen physiology especially germination and viability, has received considerable attention for its application in plants breeding, conservation, adaptation & understanding of physiological behavior of fertilizing pollen grains. There are several reports on pollen germination and viability from different taxa (Nair & Singh, 1972; Vijay, O, P; 1972; Kapoor, 1976; Zeng-Yu Wang *et al*., 2004.

Pollen grains of angiosperm can be classified into two groups, binucleate and trinucleate. The latter one loose viability very rapidly & can hardly germinate on artificial media. Pollen have considerable potential to achieve genetic transformation. There are some critical external factors which affect the maintenance of pollen germination capacity relative humidity (RH), and temperature surrounding pollen (King 1961, Gill & Malik., 1992, Malik & Thind, 1992, Shivanna & Rangaswamy, 1992). Pollen grains of different plants require varying range of growth media like water, sugar solution, inorganic salts & vitamins for successful germination. Pollen stored at low temperature presented germination capacity better than high temperature (Stanley & Linskens 1974). Khan & Perveen (2007) reported germination capacity and maintenance of *Carica papaya*. It has been widely acknowledged that temperature & relative humidity of the storage environment are two important factors, which profoundly influence the viability of stored pollen.

Pinney & Polito (1990) reported the germination of olive pollen improved markedly in storage condition. Thomas (2000) studied pollen germination of 40 plant species on sucrose gelatin on onion epidermis. According to Aslantus & Pirlak (2002) the germination capacity of strawberry pollen increase at low temperature. Germination capacity of *Morus alba* is reported by Khan & Perveen, (2008).

The present study is the first attempt to analyze storage condition & viability test method of *Ficus carica* L. No reports are available on maintenance and germination capacity of stored pollen of this economically important plant.

Material and Methods

Methodology: A polliniferous material was collected from cultivated fields of Khuzdar, Quetta & plants growing at Karachi University campus in large quantity during the peak of flowering period of species. Fresh pollen were systematically subjected to preliminary viability tests (Alexander, 1969). Pollen culture media were prepared using Brewbaker and Kwack (1963) techniques. Pollen grains equal to at least twice the diameter of pollen grains counted as germinated, burst pollen grains are not counted as germinated pollen. The viability of stored pollen was assessed in terms of percentage germination. The stored pollen was germinated in a humidity chamber in different solutions. The germination was determined after 3-6hrs of incubation. The hanging drop technique used for culturing pollen grains in liquid media, culture was stored at room temperature.

The pollen grains slides were prepared for light microscope (LM) using Erdtman (1952) procedure. Pollen grains were mounted in unstained glycerin jelly & observations were made with a Nikon type-2 microscope. The measurements are based on 15 readings.

Results and Discussions

Germination capacity of stored pollen of *Ficus carica* L. has been examined for 48 weeks in different conditions as refrigerator, freezer, vacuum and in organic solvents. Pollen stored in freezer (-30°C, -20°C) showed better germination percentage as compared to pollen stored at 4°C and in organic solvents (Table 1). Fresh and 4°C showed more or less equal germination percentage. At 4°C the germination capacity decreases as compared to germination at freezer where the germination is reasonably high after 24 weeks (Table 1). The freeze-drying condition seems to be the good method where the germination is up to 53.70% after 48 weeks (Table 1).

In organic solvents pollen grains were treated from 1-24 hrs and then stored at +4°C. Benzene showed the best germination as compared to acetone and chloroform in which pollen lost viability after 9-12 hrs. Benzene showed 11.4% germination after 18 hrs of treatment and then no germination was noted. The acetone and chloroform showed 3 to 4% germination after 12 hrs. In vacuum pollen was treated over silica jell 1-24 hrs and then germinated. This condition also showed low germination i.e., 9.6% after 18 hrs of treatment and then lost viability.

The controlled temperature & humidity conditions were found to be effective in prolonging pollen viability in *Ficus carica* L. (Fig. 1) although the extent of prolongation was highly variable between the different storing conditions.

The present investigations are the first systematic attempt to compare the efficiency of pollen of *Ficus carica* in storage through conventional methods of controlled temperature (4°C, -20°C, -30°C, -60°C) and humidity, in organic solvents. The pollen grains were fairly uniform in their response to the organic solvents tested. Germination percentage was maximum in freeze-dried pollen (Table 1)

Conclusively temperature and humidity are the major influencing factors in the pollen behavior of different conditions.

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Table 1. Germination capacity of stored pollen of *Ficus carica* L. (Moraceae) at different temperatures and humidity conditions in sucrose and boric acid solutions.

Weeks	Germination % at 4°C	% Solutions	Germination % at -20°C	% Solutions	Germination % at -30°C	% Solutions	Germination % at -60°C	% Solutions
1	52.10	20	69.00	30	71.30	30	73.12	40
2	50.80	20	70.00	30	71.00	30	71.56	40
4	50.00	30	69.20	50	70.20	40	72.10	30
8	50.00	20	66.30	60	69.00	30	70.00	30
12	47.10	30	65.10	30	67.40	30	70.10	40
16	42.60	30	62.00	30	64.40	70	69.20	40
20	40.10	40	60.10	30	62.00	30	67.00	50
24	36.70	40	56.50	30	60.00	30	64.70	40
28	32.00	20	53.00	30	58.20	40	63.00	30
32	29.50	30	50.20	40	55.60	50	61.60	30
36	25.60	30	46.15	30	56.10	30	60.00	30
40	21.00	30	42.00	30	53.10	40	58.10	40
44	18.10	30	39.70	30	51.50	40	56.00	40
48	16.00	30	35.40	20	50.00	40	53.70	60

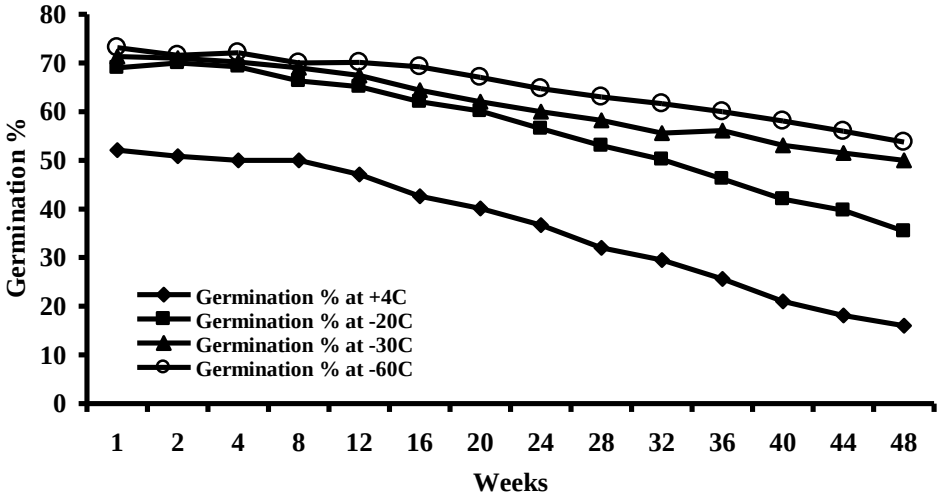

Fig. 1. Germination capacity of stored pollen of *Ficus carica* L. (Moraceae) at different temperature and humidity conditions in sucrose and boric acid solution.

References

Alexander, M.P. 1969. Different staining of aborted and non aborted pollen. *Stain Trchnology*, 44: 117-122.

Aslantus, R. and L. Pirlak. 2002. Storage of Strawberry pollen. IV International Symposium on Strawberry pollen, pollen. (Eds.): Hietaranta, M.-M Linn., Palonen & Parikka, P. Acta Hortcultureae, 2: 567.

Brewbaker, J.L. and B.H. Kwack. 1963. The essential role of calcium ion in pollen tube growth. Amer. *J. Bot*., 50: 859-865.

Erdtman, G. 1952. Pollen morphology and plant taxonomy of Angiosperms. In: *Introduction to Palynology*, 1: Almquist and Wiksell, Stockholm.

Gill, M.S. Neelam and C.P. Malik. 1992. Pollen biotechnology storage and viability. In: *Pollen Physiology and Biotechnology*. (Ed.): C.P. Malik. Today and tomorrow's Printer and Publisher, New Delhi, India.

Iwanami, Y. 1971. The soaking of pollen grains into organic solvents. *Jap. Jour. Palynol*., 8: 39-43.

Kapoor, S.K. 1976. Pollen germination in some Cucurbits. *J. Palyn*., 12:(1&2): 87-93.

Khan, S.A. and A. Perveen. 2008. Germination capacity of stored pollen of *Morus alba* L., (Moraceae) and their maintenance. *Pak. J. Bot*., 40(5): 1823-1826.

Khan, S.A. and A. Perveen. 2007. Germination capacity of stored pollen of *Carica papaya* L., (Caricaceae) and their maintenance. *Pak. J. Bot*., 39(5): 1403-1406.

King, J.R. 1961. The freeze drying of pollen. *Economic Botany*, 15: 91-98.

Malik, C.P. and S.K. Thind. 1992. Pollen biotechnology and fertilization engineering in crop improvement. In: *Pollen Physiology and Biotechnology*. (Ed.): C.P Malik. New Delhi, India.

Nair, and Singh, B.V. 1972. Pollen germination studies in some legumes. *J. Palyn*., 8: 63-68.

Pinney, K. and V.S. Polito. 1990. Olive pollen storage and in vitro germination. In: *International Symposium on Olive Growing*. (Eds.): L. Rallo, J.M. Caballero and R.S. Rscabar. ISHS Acta Horticulture: 286. Vol 1.

Shivanna, K.R. and N.S. Rangaswamy. 1992. *Pollen biology*. A laboratory manual. New Delhi. India.

Stanley, R.G. and H.F. Linskens. 1974. Pollen biology, biochemistry and management. Springer, Verlag Berlin, Heidelberg, New York.

Thomas, C.J.R. 2000. Studies on pollen germination of 40 plant species on sucrose gelatin & on Onion epidermis, Quekett. *Journal of Microscope*, 38L 463-472.

Vijay, O.P. 1972. Effect of different media on the pollen germination & growth of Cucumber pollen (*Cucumis sativa* L.). *Proc. Third. Int. Symp. Sub. Trop. & Trop*. *Hort*., Bangalore.

Zeng-Yu Wang, Yaxin Ge, Megann Scott and German Spangenberg. 2004. Viability & longevity of pollen from transgenic and non-transgenic tall fescue (*Festuca arundinacea*) (Poaceae) plants. *Am. J. Bot*., 91: 523-530.

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