Viability of Pollen Grains of *Gaillardia aristata*, Pursh, Stored in Organic Solvents

Uday. K. TIRLAPUR and M. G. PANCHAKSHARAPPA

Histochemistry and Morphogenesis Laboratory, Department of Studies in Botany, Karnatak University, Dharwad-580003, India

The pollen grains of *Gaillardia aristata*, pursh, stored in benzene, acetone, chloroform and n-heptane for over 16 months at 4°C germinated in vitro. Furthermore, the flowers pollinated by pollen grains stored in acetone and n-heptane in vivo could produce normal seeds. The results indicate that the technique of organic solvent storage can effectively be adopted for long-term storage of pollen.

Introduction

The utility of stored pollen grains has often been in overcoming physical barriers, particularly in breeding of distantly isolated and cultivated crop plants growing in different agroclimates of the world. Of late the technique of organic solvent storage of pollen seems to hold great promise for transporing pollen without dry-ice or any sophisticated instrumentation. Panchaksharappa and Tirlapur[1] earlier reported the efficacy of 25 organic solvents for preserving pollen viability/germinability in *Gaillardia aristata*, pursh, a horticultural plant. They recorded the retention of high degree of germinability of pollen soaked in organic solvents for 33hours following storage at 4°C Their observations were basically similar with the studies of Iwanami.[2-4] However, the retention of pollen viability in organic solvents for more than 1 year has usually been at subzero temperature[5,6] and there have been no reported instances of long-term retention of pollen viability in organic solvents maintained at temperature above 0°C

The present communication reports for the first time the feasibility of effectively storing pollen grains in organic solvents at 4°C over a period of 16 months.

Materials and Methods

The procedure for storing the pollen grains in organic solvents is same as described earlier.[1]

The grains were stored in organic solvents in March 1984 and after 16 months of storrage

at 4°C the pollen grains of *Gaillardia ariatata* were taken out from the solvents in July 1985, by mean of a dropper. They were then filtered and dessicated with an aspir-ator to remove the trace of solvent from surface of the pollen grains. Around 500 pollen grains were collected from the filter paper and cultured on medium containing 20% sucrose, 2% agar, 50 ppm of boric acid and 100 ppm of $CaCl_2$. The cavity slides containing the culture medium with pollen grains were maintained at 95% of humidity initially and then at 40% humidity for 7 hours. Of the two replicates 400 pollen grains were scored to record the percentage germination. Pollen grains retained at room temperature under uncontrolled conditions and those freshly collected were also cultured on separate cavity slides to compare the percentage germination with those of the stored ones.

Results and Discussion

The percentage germination of pollen grains stored in organic solvents, those retained under uncontrolled conditions and grains freshly collected from the plants growing in the departmental garden are presented in Table 1.

The grains stored at room temperature for 16 months(control) and those stored in petroleum ether and ether did not germinated at all. However, it was interesting to note that pollen soaked in benzene, acetone, chloroform and n-heptane had retained their viability and showed 52-66% germinability at the end of 16 months of storage with an average pollen tube length of 7-8 mm.

The grains stored in n-heptane and acetone were used to pollinate the flowers in vivo. It was observed that by mid September 1985, the flowers pollinated in vivo by such stored

Table 1. Germinability of pollen grains stored in 6 organic solvents at 4°C for 16 months.

Organic solvents	Percentage of germination
Petroleum ether	0
Ether	0
Benzene	52.3
Acetone	59.2
Chloroform	47.4
n-Heptane	65.9
Control (unsoaked)	0
Freshly collected pollen	98.9

pollen had set 45-57 seeds per flower of which on an average 38 seeds collected from each flower were viable and they could be germinated into healthy seedlings.

It is therfore obviously clear from the present study that the technique of storing pollen in organic solvents particularly in n-heptane and acetone holds promise and potential in long-term storage at 4°C. In view of the simplicity of this technique over others requiring freezed-drying, ultralow temperature and vacuum systems for pollen storage it can be concluded from the present study that pollen soaked in organic solvents can effectively be used in breeding programmes and in germ plasm storage.

Further work is in progress in this laboratory to study the efficacy of this technique to store pollen grains of a wide variety of economically important taxa.

References

(1) Panchaksharappa, M. G. and Uday K. Tirlapur: J. Palynol. **20**, 132-134 (1984).
(2) Iwanami, Y.: Botanique **3**, 61-68 (1972).
(3) Iwanami, Y.: Plant and Cell Physiol. **13**, 1139-1141 (1972).
(4) Iwanami, Y.: Protoplasma **84**, 181-184 (1975).
(5) Iwanami, Y. and N. Nakamura : Stain technol. **47**, 137-139 (1972).
(6) Iwanami, Y.: Experientia **40**, 568-569 (1984).

(Accepted May 2, 1986)