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A Note on Effect of Storage of Date Palm (*Phoenix dactylifera* L.) Pollen Grains on Their Germination and Date's Fruit Setting

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Abstract: This investigation was initiated to study the effect of different storage methods on *in vitro* germination of date palm pollen grains as well as on fruit-setting. The collected pollen grains were stored in a refrigerator, freezer and freezer after being immersed in liquid nitrogen. After one year of storage, pollen viability was evaluated in terms of *in vitro* germination percentage and fruit-setting. The results revealed that the highest germination percentage and fruit-setting were associated with fresh pollen grains stored in the freezer followed by storage in freezer after immersion in liquid nitrogen and storage in the refrigerator. These results indicated that well dried pollen grains could be stored in fairly viable state until the following season in a freezer or a refrigerator.

Key words: Date palm; pollen storage; pollen germination; fruit set

Date palm is a dioecious crop, and artificial pollination is required to obtain satisfactory fruit setting (Zaid 1999). Pollen grains have direct effect on fruit characters in addition to the time of ripening. The early emergence of inflorescences on female palm trees before opening of adequate number of male spathes necessitates storage of pollen during the pollination period or from season to season. Recently, thousands of fruiting palms were planted in Sudan through introduction of tissue-cultured seedlings. No tissue – cultured male palm trees are available. Farmers rely on local males for pollination. This may lead to the spread of

pests and diseases to the valuable female palms. Thus, availability of high quality pollen and their efficient storage are of paramount importance to date palm growers. The aim of this study was to develop simple reliable methods for the storage of date palm pollen.

Male spathes were collected from male palms at Khartoum North, Khartoum State**.** Each healthy mature spathe, free from insects, was opened and strands dried on metal mesh at room temperature. Dried pollen grains were collected after shaking the strand on a sheet of paper. Pollen grains were subjected to further drying in a desicator using silica gel. Dehydrated pollen grains of two males were used; namely, Male 1 (M1) from Shabia, Khartoum North and Male 2 (M2) from Shambat, Khartoum North. Pollen grains from M1 and M2 were stored in closed containers in a refrigerator, freezer, and freezer after immersion in liquid nitrogen. Small quantities of pollen grains were treated with liquid nitrogen by wrapping them in a piece of aluminum foil and then immersing them in liquid nitrogen. Treated pollen were kept in closed vials and stored in the freezer.

For *in vitro* pollen germination, a medium with 15% sucrose, 200 mg/l $CaNO_3$, 50 mg/l boric acid and 150 mg/l KNO_3, solidified with 1% agar, was used (Mortazvi *et al.* 2010). The medium was dispensed into petri dishes (90 mm diameter). Pollens were sown using a piece of cotton. Incubation was at room temperature. After 8 hrs incubation, observations were made by counting 300 pollens under 40 X magnification using CETI microscope (Fig.1). *In vivo* pollen viability was determined by estimating fruit set. Pollination for fruit set tests was carried out using cotton balls dusted with pollen. Bunches were covered with paper bags upon pollination.

Male 1 was used for pollen germination test. Pollen grains from Male 1 were used in preliminary trials to select a suitable medium for *in vitro* germination test. Amounts of pollen remained for *in vivo* germination test were not enough. Thus, Male 2 was used in pollination of Sagge date palm, grown at Soba, Khartoum, to test fruit setting. Two palm trees were used; Four bunches in each individual tree were pollinated, each with one pollen source (treatments). Each palm was considered a block. Fruit set percentage was an average in two bunches, one from each palm. Ten

strands from each bunch were used for estimation of fruit setting. The data were analyzed as randomized complete block design. The statistical package Genstat (2011) was used to run the analysis.

The highest germination percent was obtained with storage of pollen in the freezer (26.0%) followed by deep freezer (22.6%), liquid nitrogen (18.0%) and the lowest by refrigerated storage (14.3%) (Table 1). These results are in accord with those of Mortazavi *et al*. (2010) who reported higher pollen germination with pollen stored in the freezer compared to refrigerator.

Table 1. *In vitro* pollen germination and fruit set percentages after one year of storage at different storage conditions

Storage conditions	In vitro pollen germination percentage	Mean% fruit set
Fresh	26.0	62.6
Freezer	22.6	53.3
Freezer/Liquid nitrogen	18.0	46.5
Refrigerator	14.3	45.6
Mean		52.0
SE ±		4.84

Fruit set is the targeted goal of pollination. Pollination with pollen stored under different conditions was performed on Sagge female date palm, in El-Nefadi Farm, Soba, Khartoum and the results indicated satisfactory fruit setting after one year of storage under all storage conditions (Table 1; Fig.1). Fresh pollen resulted in the highest fruit set (62.60 %) followed by pollen stored in the freezer (53.3%), pollen stored in the freezer after immersion in liquid nitrogen (46.5%) and from pollen stored in the refrigerator (45.6%). No significant difference, in fruit set percentage was found between fresh pollen and pollen stored in the freezer (Table 1). There was a significant difference between fresh pollen and pollen stored in the freezer and in the freezer after immersion in liquid nitrogen and storage in the refrigerator. Storage of pollen in the freezer after emersion in liquid nitrogen was expected to give significantly higher viability than storage in the freezer (Mortazavi *et al*. 2010), but, our results indicated slightly lower, yet non-significant viability of pollen stored in the freezer

after immersion in liquid nitrogen compared to storage in the freezer (Table1).

The higher fruit set percentage compared to germination test indicated that the germination test underestimated the viability of pollen under the test conditions used in this study. This can be explained by the fact that all the conditions for optimum pollen germination were not provided, particularly incubation temperature and pH of the culture medium (Smail and Zohair 2013; Smail 2014). Based on the results obtained, date palm pollen could be stored in fairly viable state until the following season under refrigeration or freezer temperature conditions after drying using silica gel.

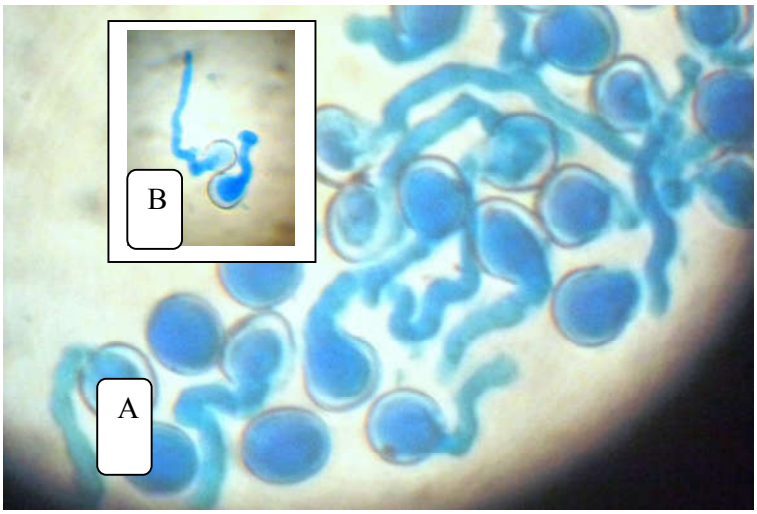

Fig.1.A: Germination test of pollen from Male 2 stored under freezer conditions for one year; B: close view of pollen germination

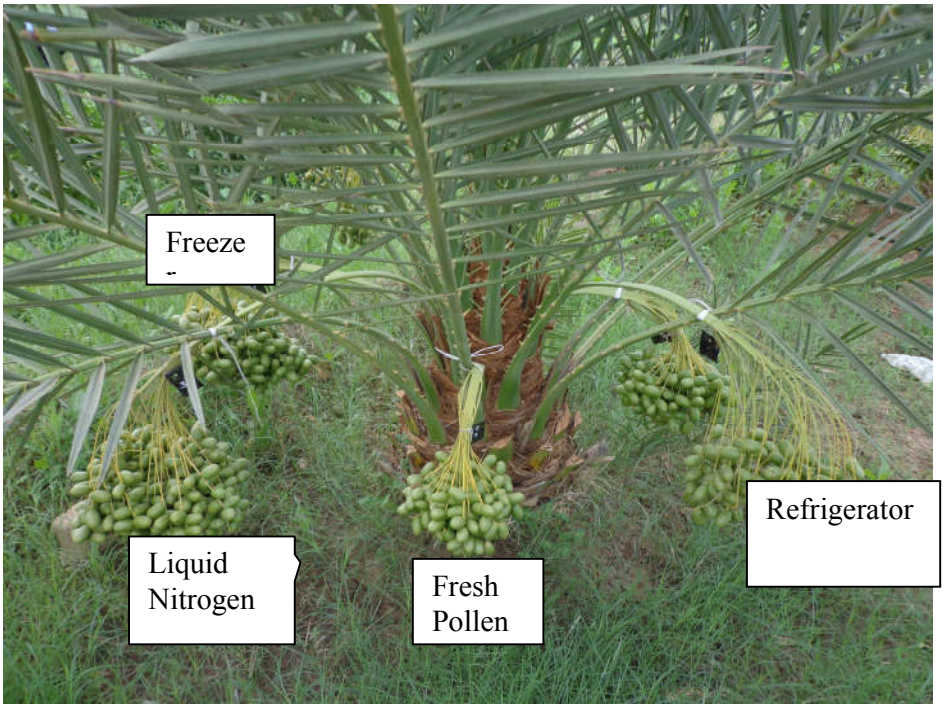

Fig. 2. Sagge date palm pollinated with fresh pollen, pollen stored at the refrigerator, freezer and liquid nitrogen

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تأثير التخزين على إنبات حبوب اللقاح وعقد الثمار فى النخيل

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المستخلص: أُجري هذا البحث لدراسة تأثير طرائق تخزين مختلفة على إنبات حبوب لقاح شجرة نخيل التمر . خزنت حبوب اللقاح فى الثلاجة والمجمد والمجمد بعد غمرها في النيتروجين السائل . بعد عام من التخزين تم تحديد حيوية حبوب اللقاح في المختبر بإختبار النسبة المئوية لإنباتها . كما تم تحديد حيوية حبوب اللقاح في الحقل بحساب النسبة المئوية لعقد الثمار .
أعلى نسبة إنبات تم الحصول عليها من حبوب اللقاح الطازجة (26.0%) تليها نسبة الإنبات في التخزين في المجمد (26.6%) تليها نسبة الإنبات في التخزين في المجمد بعد الغمر في النيتروجين (18.0%) ثم حبوب اللقاح المخزنة في الثلاجة (14.3%) . أعلى نسبة عقد للثمار سجلت عند استخدام حبوب اللقاح الطازجة (62.6%) تليها نسبة عقد للثمار من حبوب اللقاح المخزنة في المجمد (53.3%) ثم نسبة عقد من حبوب اللقاح المخزنة في المجمد بعد الغمر في النيتروجين (46.5%) . وأخيراً نسبة عقد للثمار من حبوب اللقاح المخزنة في الثلاجة (45.5%) . بينت الدراسة أن من الممكن تخزين حبوب اللقاح فى المجمد أو الثلاجة والإحتفاظ بها حية بنسبة معقولة .