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Proper Environment Improves the Storage of Primed Pansy Seed

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Abstract. **Primed and nonprimed 'Majestic Giant *Yellow*' pansy *(viola* x *wittrockiana* Gams.) seed had similar total germination percentages, but priming reduced the number of days to 50% of final germination by 25% and the days between 10% and 90% germination by 21%. Reducing the moisture content of nonprimed seed from 10.5% to 5.8% caused no change in total germination, but primed seed germination decreased at moisture contents below 10%. Total germination percentages of primed and nonprimed seeds declined as storage temperatures were reduced from 5 to - 20C. Storing primed seed at SC and 11% relative humidity (RR) for 16 weeks reduced seed moisture contents to 8.4% and total germination by 28%. Primed seeds, after 5C storage at 75% or 95% RH for 16 weeks, contained 23.7% and 28.3% moisture, and germination had declined 31% and 48%, respectively. Primed seed stored 16 weeks at 5C and 52% RR had 12.5% seed moisture content and germination had declined by 2%. Moisture contents of primed pansy seeds must remain between 12% and 20% during 5C storage or total germination will decline and the benefits of more rapid and uniform germination are lost.**

Seed priming, also termed osmotic priming or osmoconditioning, limits the water seeds imbibe, permitting the initial stages of germination, but inhibiting radicle penetration of the testa (Heydecker, 1977). Priming promotes faster and more uniform seedling emergence, even at unfavorable temperatures (Cantliffe, 1981). Priming seeds in osmotic solutions of polyethylene glycol (PEG 8000) or potassium salts K_3PO_4 and KNO_3 promoted more rapid and uniform germination of tomato *(Lycopersicon esculentum* Mill) (Bradford and Murray, 1983), lettuce *(Lactuca sativa* L.) (Cantliffe, 1981), pansy (Carpenter and Boucher, 1991), and salvia *(Salvia splendens* F. Sellow ex Roem. & Schult) (Carpenter, 1989) seeds. For many species, the advantages of seed priming were diminished or lost when seeds were stored (Heydecker, 1977), which has been attributed to embryo desiccation during storage (Bewley and Black, 1985). Carpenter (1989, 1990) reported that primed salvia seeds stored at 5C for 1 to 16 weeks had reduced total germination and lost the capacity for earlier germination. Primed dusty miller *(Senecio cineraria* DC.) seed germination, however, was unchanged after 16 weeks of storage at 5C and 52% relative humidity (RI-I). The research objectives of these experiments were to determine the changes in rate, uniformity, and total germination of primed pansy seed following low-temperature seed treatment, desiccation to low seed moisture levels, or after storage at various RH levels.

Seed priming. 'Majestic Giant Yellow' pansy seeds were primed in aerated PEG 8000 solutions at -1.0 MPa for 7 days at 15C (Carpenter, 1991). Water potentials of PEG 8000 solutions were measured with a Wescor vapor-pressure osmometer (Wescor Co., Logan, Utah) at 15C. Following priming, seeds were washed with 100 ml of distilled water (DW), surface-dried, and stored 1 day at 5C and 45% RH. Primed and nonprimed treatments contained four 100-seed replications. Seeds were germinated at 20C in g-cm petri dishes on two Whatman No. 1 filter papers moistened with 5 ml of DW. Germinated seeds with visible radicle protrusion through the testa were counted daily. Total germination percent (G), number of days to 50% of final germination (T_{50}), and germination span in days between 10% and 90% germination (T_{90} - T_{10}) were calculated (Furutani et al., 1985). The design was a randomized complete block, with data for G, T_{50}, and rate of increase in germination (K) of primed and nonprimed seeds analyzed by nonlinear regression procedures using SAS (1985) PROC NLIN, with estimated values compared using the asymptotic 95% confidence intervals.

Reducing primed seed moisture content. Seeds were weighed after priming, preparation, and storage as described above. The four 100-seed replications comprising each treatment were placed in open 9-cm petri dishes and dehydrated for 0, 4, 8, 12, 24, 48,72, or 96 h at 35C in forced-draft ovens. Seeds were reweighed immediately after they had cooled to ambient temperature and then were sealed in screw-capped 4-ml glass vials, 100 seed per vial, before being stored at 5C for 6 weeks. Seeds were reweighed after storage, germinated at 20C as described above, and germination counts were made daily. Total moisture content of primed and nonprimed seeds was determined by weighing four replications before and after 48 h desiccation in a forced-draft oven at 105C. The design was a randomized complete block, with data analyzed by analysis of variance (ANOVA) and polynomial regression to investigate the germination trend resulting from reduced seed moisture content.

Temperature effects on primed seeds. Seeds primed for 1 week were washed and surface moisture was removed by vacuum aspiration for 30 min. Primed and nonprimed seeds, consisting of four l00-seed replicates, were placed in 15 x 2.5-cm petri dishes on wire screens supported by segments of plastic tubing 1 cm above a chemical desiccant. Constant RH (34%) was maintained in the dishes sealed with parafilm after 50 ml of saturated magnesium chloride was added to

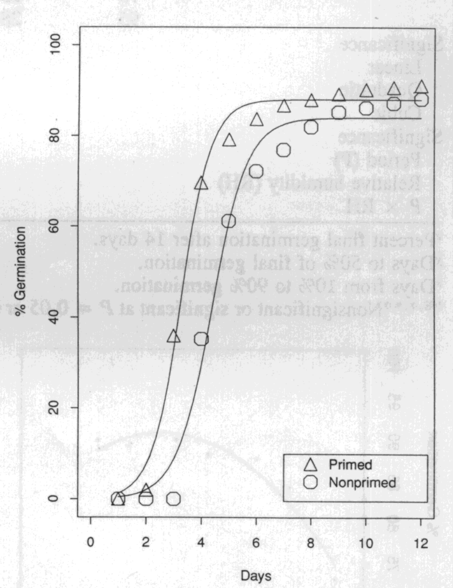

Fig. 1. Cumulative germination of seeds in incubators at constant 20C, comparing seed primed with - 1.0 MPa PEG 8000 at 15C for 7 days with nonprimed seed of 'Majestic Giant Yellow' pansy. The estimated values for total germination percentage (G), days to 50% of final germination (T_{50}), and rate of increase (K) were obtained with the equation: % germination = $G/l + e^{-k(day - T50)}$ (Ratkowsky, 1990), and compared for primed and nonprimed seeds using asymptotic 95% confidence intervals.

$\hat{G}$	Primed	88*
	Nonprimed	84
$\hat{T}_{50}$	Primed	3.3*
	Nonprimed	4.4
$\hat{K}$	Primed	1.8^{NS}
	Nonprimed	1.5

*95% confidence intervals do not overlap, indicating significance at $P \le 0.05$.

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Table 1. Germination of primed pansy seed after storage at various relative humidities.

Seed storage			Germination		
Period (weeks)	RH (%)	Seed moisture (%)	G^z (%)	$T_{50}{}^y$ (days)	$T_{90} - T_{10}{}^x$ (days)
0		20.7	89	3.3	3.1
2	11	13.6	90	3.4	3.6
	52	15.2	89	3.3	3.2
	75	19.5	89	3.2	2.9
	95	22.2	91	3.3	2.8
Significance					
Linear			NS	NS	NS
Quadratic			NS	NS	NS
Cubic			NS	NS	NS
4	11	10.1	88	3.7	4.4
	52	13.9	89	3.3	3.5
	75	21.0	89	3.2	3.2
	95	24.6	67	3.4	3.4
Significance					
Linear			*	NS	NS
Quadratic			**	NS	NS
Cubic			**	NS	NS
8	11	8.9	74	3.9	4.3
	52	13.2	89	3.3	3.4
	75	21.8	78	4.0	3.9
	95	29.7	62	4.5	3.8
Significance					
Linear			NS	NS	NS
Quadratic			**	**	**
Cubic			*	NS	NS
16	11	8.4	65	4.2	4.9
	52	12.5	88	3.4	3.6
	75	23.7	62	4.1	4.3
	95	28.3	47	4.8	4.2
Significance					
Linear			NS	NS	NS
Quadratic			**	**	**
Cubic			**	NS	NS
Significance					
Period (P)			**	**	**
Relative humidity (RH)			**	**	**
P × RH			**	**	*

[z] Percent final germination after 14 days.
[y] Days to 50% of final germination.
[x] Days from 10% to 90% germination.
NS,*,** Nonsignificant or significant at $P = 0.05$ or 0.01, respectively; data are the means of 400 seeds.

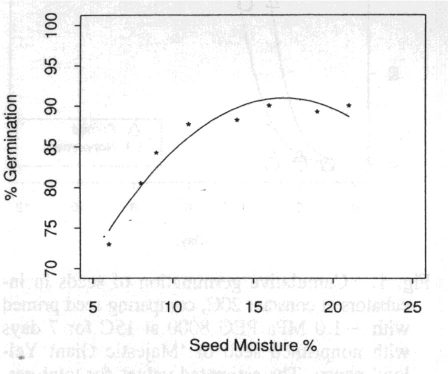

Fig. 2. Total germination percentages of primed 'Majestic Giant Yellow' pansy seeds after dehydration and storage for 6 weeks at reduced seed moisture (SM) contents. Regression equation: percent germination = $51.97 + 4.61 \times \text{SM\%} - 0.14 \times (\text{SM\%})^2$ ($R^2 = 0.85$).

the bottom of each dish (Copeland, 1976). Refrigerated incubators were maintained at 15C for 1 week to reduce seed hydration, then seeds were weighed. Immediately after weighing, the seeds were placed in l0-ml glass vials, sealed with screw caps, and immersed in polyethylene glycol-water (v/v) in controlled-temperature baths (Guy and Carter, 1984) and held for 7 days at 5,0, -5, - 10, - 15, or -20C. Bath temperatures were lowered 3C/h to final temperatures, then increased 4C/h to 1OC at the end of the 7 days. Following the temperature treatments, seeds were germinated in 20C incubators as described above. Germination counts were made daily of seeds with radicle protrusion through the testa. The design was a 2 **x** 6 factorial arranged in a randomized block, with data analyzed by an ANOVA and the trend in germination due to temperature investigated using polynomial regression.

Relative humidity during seed storage. Seeds primed for 1 week were prepared as previously described, dusted with 3a,4,7,7a-tetrahydro-2-[(trichlorome-thyl)thio]-1H-iso-indole-1,3(2H)-dione (captan), and stored at 5C and 11%, 52%, 75%, or 95% RH for 2, 4, 8, or 16 weeks; humidity treatments were achieved using saturated lithium chloride, magnesium nitrate, sodium chloride, or potassium nitrate, respectively, as described above. Incubators (model CB-1; Stults Scientific Engineering Corp., Springfield, Ill.) maintained constant temperatures during seed storage or germination. Four l00-seed replications were germinated at 20C, as described previously, immediately following each storage duration and RH level, while another four replications were desiccated 48 h in forced-draft ovens at 105C to determine stored seed moisture content. The design was a 4 **x** 4 factorial arranged in a randomized complete block, with data analyzed by ANOVA and polynomial regression analyses to determine primed seed storage duration in relation to storage RH.

Primed and nonprimed pansy seeds had similar germination curves, but primed seed had higher germination rates and required fewer days to T_{50} (3.3 vs. 4.4 days) (Fig. 1). Reduction of T_{50} by 1 day, or 25%, permitted more rapid and uniform germination. This difference could be important for commercial producers seeding directly in plug trays. There were no differences in the rates of increase during the germination of primed and nonprimed seeds (Fig. 1). Heydecker (1977) reported priming limited the water imbibition, which permitted all seeds to complete the initial stages of germination, promoting faster and more uniform seedling emergence.

Primed pansy seed tolerated only limited desiccation. Seeds dehydrated to 9% moisture content or lower and stored for 6 weeks had reduced total germination (Fig. 2). Total germination was similar for primed seeds stored at 12% to 20% moisture contents (Fig. 2). Nonprimed seeds were very tolerant of desiccation, and storage at seed moisture contents from 10.5% to 5.8% for 6 weeks failed to change germination rates (data not presented). These results support the conclusions of Bewley and Black (1985), Heydecker (1977), and Heydecker and Wainwright (1976), who found that the advantages of priming are lost or reduced if primed seeds are dried before sowing.

Primed and nonprimed seeds had similar germination rates following storage at all low temperature treatments (Fig. 3A). However, total germination percentages declined linearly for both primed and nonprimed seeds as storage temperatures were reduced from 5 to -20C. Combining the data for both seed treatments before conducting a simple linear regression analysis developed the trend line shown in Fig. 3A. These results show that significant reduction in germination percentages occur when pansy seeds are stored at excessively low temperatures. Primed seeds stored 1 week between 5 and - 20C continued to require fewer days to T_{50} than nonprimed seeds and had fewer days from $T_{90} - T_{10}$ (Fig. 3 B and C). The earlier and more uniform germination of primed than of nonprimed seed continued after storage at all temperatures.

Seed moisture content depended on RH during storage of seed at 5C, and RH governed the storability of primed pansy seed (Table 1). Storing primed seeds at 11% RH for longer than 4 weeks caused a significant decline in germination rates. The 8.4% seed moisture content after 16 weeks storage at 11% RH was within the range found for re-

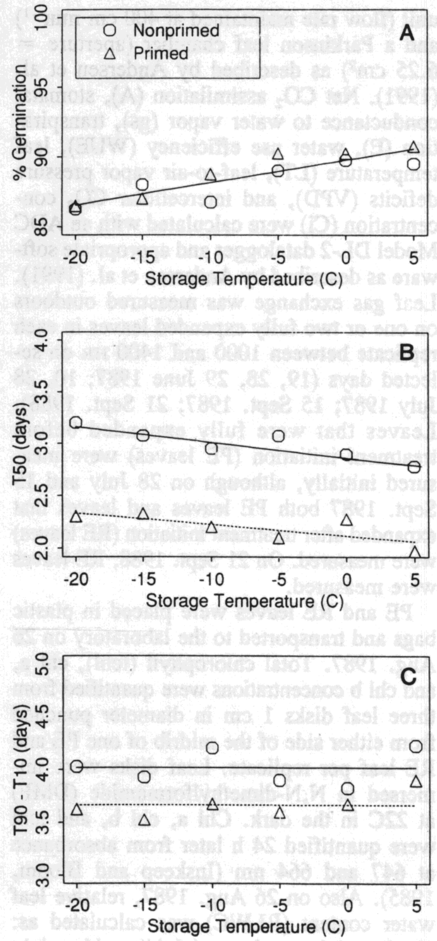

Fig. 3. Regression equations for **(A)** total germination percentages, **(B)** days to 50% of final germination, and **(C)** days between 10% and 90% germination were determined for primed and nonprimed 'Majestic Giant Yellow' pansy seed germinated after 7 days storage at 5 to −20C. **(A)** primed and nonprimed: $\hat{G} = 89.8 + 1.61 \times \text{temp.}$ ($R^2 = 0.24$); **(B)** primed: $\hat{T}_{50} = 2.12 - 0.011 \times \text{temp.}$ ($R^2 = 0.14$) ($P \leq 0.05$ or nonsignificant), nonprimed: $\hat{T}_{50} = 2.92 - 0.012 \times \text{temp.}$ ($R^2 = 0.12$); **(C)** primed: $\overline{T_{90} - T_{10}} = 4.02$ ($P \leq 0.01$), nonprimed: $\overline{T_{90} - T_{10}} = 3.67$.

duced germination-when seeds were desiccated (Fig. 2). Moisture contents of primed seeds stored at 75% and 95% RH were 23.7% and 28.3%, respectively, which likely contributed to reduced germination after storage periods of 8 or 16 weeks (Table 1). Storing primed seeds for 16 weeks at 5C and 52% RH resulted in 12.5% seed moisture and reduced germination only 2%. Further research is needed to determine the interaction of temperature and RH in storing primed pansy seed and to more accurately determine the optimal RH range. The earlier T_{50} and shorter $T_{90} - T_{10}$ of primed seed were lost after storage at 11%, 75%, or 95% RH for 4 weeks or longer (Table 1). Moisture content of primed pansy seeds must remain between 12% and 20% during 5C storage for the benefits of priming to persist. Reduced performance of primed pansy seed was found after storage at moisture contents of 8.4%, 23.7%, or 28.3%. The adoption of seed priming for pansy, and other species, will depend on the successful long-term storage of primed seed.

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