

Hydropriming of differing sized impatiens 'Expo Wine' seeds

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Summary

Priming effects may be different among impatiens seed lots with different seed sizes. To investigate this relationship, six seed size classes of 'Expo Wine' impatiens seeds were hydroprimed for different durations followed by slow dehydration using saturated salt solutions. Germination results showed that 24, 36 and 42h of hydropriming reduced speed of germination, while 4 and 8h of hydropriming promoted speed of germination and 1h hydropriming showed no significant difference compared to non-primed seeds. Statistical analysis indicated that seed size effects existed, with larger seeds germinating faster than smaller seeds following hydropriming. These results indicate that seed lots subjected to hydropriming must be sized to obtain a uniform priming response.

Introduction

The relationships between seed size and germination response have been studied for wheat (*Triticum aestivum* L.) (Grieve and Francois, 1992), onion (*Allium cepa* L.) (Gamiely, Smittle and Mills, 1990) and alfalfa (*Medicago sativa* L.) (Townsend, 1992) seeds. Studies on the relationship between wheat seed size and yield have provided conflicting results. Several investigators found that the advantages derived from large initial seed size were limited to the early stages of growth and did not contribute to increased wheat yield (Mian and Nafziger, 1992). Lafond and Baker (1986a) found that small wheat seeds not only germinated faster than large seeds, but that their seedlings also emerged more quickly (Lafond and Baker, 1986b). In contrast, the use of large seeds has frequently been shown to improve stands with more vigorous plants that significantly out performed plants derived from small seeds (Hampton, 1981).

However, to our knowledge, no studies have been reported on the affect of seed size on priming response. It is known that environmental factors such as prolonged droughts (Meckel *et al.*, 1984), high temperature (Shimzu *et al.*, 1979), light (Gray *et al.*, 1986) and position of the seed in the inflorescence (Simmons and Crookston, 1979) affect seed size. Different seed sizes might also indicate different maturation levels which may influence seed quality levels. Welbaum and Bradford (1991) reported that the success of osmopriming was dependent on muskmelon (*Cucumis melo* L.) seed maturity. Seeds harvested 40 days after anthesis (DAA) were more responsive to osmopriming than seeds harvested 60 DAA. Seed vigor can influence seed priming response. Perkins-Veazie and Cantliffe (1984) reported that priming alleviated thermodormancy of nonaged seeds of

'Great Lakes' lettuce (*Lactuca sativa* L.), but not for artificially aged seeds. It is also hypothesized that since smaller seeds have a higher ratio of surface area/volume compared to larger seeds, they are normally hydrated more rapidly than larger seeds (Muchena and Grogan, 1972) which may influence priming response. This study was designed to investigate whether different size classes of impatiens seed affect priming response.

Material and methods

Seed material

Six size classes of 'Expo Wine' impatiens seeds ranging from 1.0 to 1.5 mm diameter were provided by PanAmerican Seed Company in December 2001. Seeds were stored in foil bags in sealed desiccators at 4°C during the course of the study.

Hydropriming method

Seeds were placed on two layers of water-saturated blotter paper (Anchor Paper Co., St. Paul, MN) in an 11 × 11 × 4 cm plastic box. Seeds were positioned in a monolayer to ensure that sufficient air surrounded the seeds. Hydropriming occurred for 1, 4, 8, 24, 36 and 42h under light and at room temperature (25 ± 1°C).

Dehydration after priming

Following hydropriming, seeds were placed on top of a metal screen inside a sealed plastic box, 50 ml of saturated salt (NaCl) solution was added to the bottom of the box (which provided a relative humidity of 75%) and the salt did not contact the screen or seeds. The seeds were dried in the sealed box for 8 h, then another 3 h were required to dry the seeds at ambient room conditions (25 ± 1°C) in order to bring the seeds to their approximate original moisture content (8% fwb).

Germination test

Germination conditions were according to the ISTA Rules for Testing Seeds (1999). Fifty seeds were placed in covered Petri dishes containing two layers of blotters moistened with 9 ml distilled water and germinated at 25°C and 85% RH with 8 h fluorescent light and 16 h dark. Seeds were identified as germinated when the radicle protruded and a first count was taken at day 7 and a final count at day 18. Mean time to germination (MTG) values were calculated by using the following formula:

$$\text{MTG (in days)} = ((N1 * T1 + (N2 - N1) * T2 + (N3 - N2) * T3 + \dots + (N7 - N6) * T7)) / N7$$

In which: N = % germination of the seeds

T = days

$$\text{MTG (in hours)} = \text{MTG (in days)} * 24$$

Fresh weight

The average fresh weight of five classes of 'Expo Wine' impatiens seeds was determined for 200 seeds as: Seed 1 1043 mg, Seed 2 1263 mg, Seed 3 1561 mg, Seed 4 1637 mg and Seed 5 1922 mg. Fifty seeds of each size category were then hydroprimed as described above for 1, 2, 4, 6 and 8 h. Following the hydration period, the fresh weight was determined and the percentage fresh weight increase over time recorded.

Results and discussion

Larger, non-primed seeds germinated faster and produced heavier seedling fresh weights (tables 1 and 2). All size classes of non-primed seeds had 100% germination by day 7 (figure 1). When priming for 24, 36 and 42h, 'Expo Wine' impatiens seeds showed reduced germination speed compared to non-primed seeds. After 4 and 8h of hydropriming, speed of germination increased while 1h hydropriming had no significant effect compared to non-primed seeds (table 1). When primed for 4 and 8 h, speed of germination was promoted, but this faster germination was not translated into greater seedling fresh weight except for the two larger seed size classes at 4h priming (table 2).

Table 1. Mean time to germination (MTG) following hydropriming 0, 1, 4, 8, 24, 36, or 42 h for six different size classes of impatiens 'Expo Wine' seed. **A** indicates seed diameter > 1.5mm; **B** indicates seed diameter between 1.4 and 1.5mm; **C** indicates seed diameter between 1.3 and 1.4 mm; **D** indicates seed diameter between 1.2 and 1.3 mm; **E** indicates seed diameter between 1.1 and 1.2 mm, **F** indicates seed diameter < 1.1mm. The individual means are for 150 seeds.

Seed Size	Priming Time (h)						
	0	1	4	8	24	36	42
	MTG (h)						
A	61.7 (0.9)	61.2 (2.2)	55.2 (1.3)	55.9 (2.4)	67.9 (1.1)	68.7 (1.7)	69.1 (2.6)
B	65.5 (1.0)	64.3 (3.1)	60.5 (1.0)	62.4 (0.6)	74.6 (3.2)	74.9 (1.7)	71.8 (4.6)
C	67.2 (2.1)	65.8 (1.3)	63.6 (1.4)	64.1 (3.4)	76.6 (3.4)	76.3 (3.1)	73.2 (3.8)
D	70.3 (1.0)	69.8 (3.9)	67.4 (2.4)	69.1 (2.1)	78.5 (3.1)	78.5 (2.4)	78 (4.8)
E	70.3 (1.2)	70.1 (1.2)	67.4 (2.7)	68.6 (1.2)	79.2 (1.3)	79.6 (1.9)	81.6 (5.0)
F	72.2 (0.7)	72.9 (1.8)	70.6 (3.0)	72 (1.5)	82.3 (2.6)	82.8 (1.9)	84.2 (6.2)

* The standard error for each mean is indicated in parentheses.

Table 2. Seedling fresh weight (50 seedlings) following hydropriming 0, 1, 4, 8, 24, 36, or 42 h for six different size classes of impatiens 'Expo Wine' seed. **A** indicates seed diameter > 1.5mm; **B** indicates seed diameter between 1.4 and 1.5mm; **C** indicates seed diameter between 1.3 and 1.4 mm; **D** indicates seed diameter between 1.2 and 1.3 mm; **E** indicates seed diameter between 1.1 and 1.2 mm, **F** indicates seed diameter < 1.1mm. The individual means are for 150 seeds.

Seed Size	Priming Time (h)						
	0	1	4	8	24	36	42
	Fresh weight (g)						
A	0.50 (0.04)	0.52 (0)	0.55 (0.02)	0.48 (0.04)	0.47 (0.03)	0.41 (0.05)	0.37 (0.02)
B	0.46 (0)	0.45 (0.02)	0.49 (0.02)	0.41 (0.03)	0.34 (0.04)	0.35 (0.03)	0.33 (0.04)
C	0.44 (0.01)	0.43 (0.02)	0.43 (0.06)	0.36 (0.01)	0.35 (0.01)	0.35 (0.04)	0.30 (0.01)
D	0.38 (0.02)	0.37 (0.04)	0.36 (0.02)	0.31 (0.01)	0.31 (0)	0.30 (0.01)	0.27 (0.02)
E	0.32 (0.01)	0.33 (0.01)	0.34 (0.02)	0.30 (0.01)	0.26 (0.02)	0.27 (0.01)	0.23 (0.03)
F	0.26 (0.01)	0.31 (0.01)	0.31 (0.01)	0.27 (0.02)	0.22 (0.01)	0.23 (0.02)	0.20 (0.03)

* The standard error for each mean is indicated in parentheses.

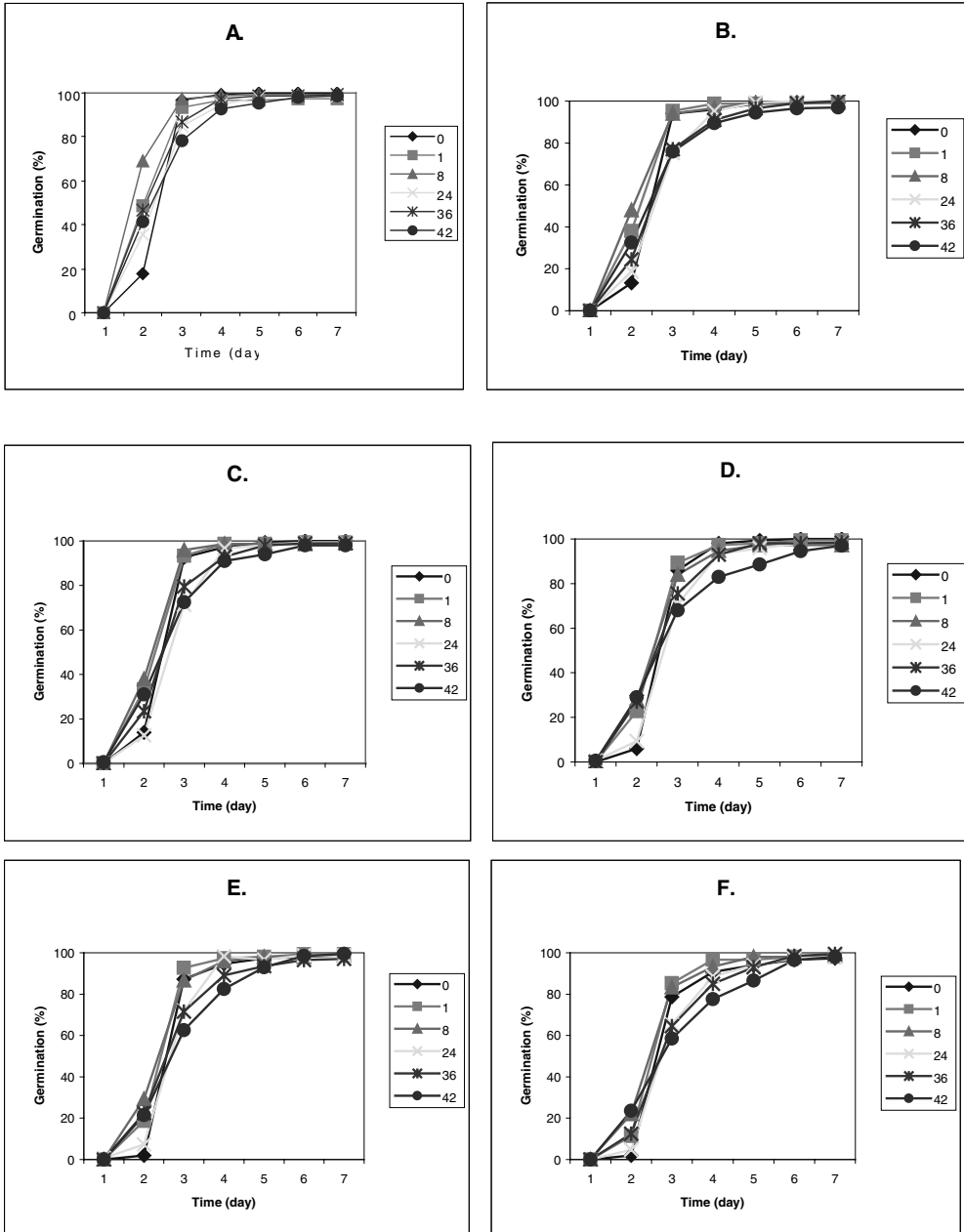


Figure 1. Effect of seed hydropriming treatments (0, 1, 4, 8, 24, 36, or 42 h) for six different seed size classes in relation to percent germination at 25C of 'Expo Wine' impatiens. The individual means are from 150 seeds. **A** indicates seed diameter > 1.5mm; **B** indicates seed diameter between 1.4 and 1.5mm; **C** indicates seed diameter between 1.3 and 1.4 mm; **D** indicates seed diameter between 1.2 and 1.3 mm; **E** indicates seed diameter between 1.1 and 1.2 mm, **F** indicates seed diameter < 1.1mm.

To investigate the relationship among seed size classes and priming, MTG values were analyzed as a factorial experiment with three replications using SAS protocol of general linear model (GLM). Main factors were hydropriming time (0, 1, 4, 8, 24, 36, or 42h) and seed size (diameter of seed >1.5mm, 1.4-1.5mm, 1.3-1.4mm, 1.2-1.3mm, 1.1-1.2mm, <1.1mm). Priming durations of 24, 36 or 42h priming time delayed 'Expo Wine' impatiens seed germination while 4 or 8h of hydropriming promoted germination (table 3). Hydropriming for only 1h had no effect (table 3). Seed size effects also existed with larger impatiens seeds germinating faster than smaller seeds after hydropriming (table 4).

Table 3. Seed hydropriming treatments (0, 1, 4, 8, 24, 36, or 42 h) for six different seed size classes in relation to the mean time to germination (MTG) and seedling fresh weight (50 seedlings) on the 7th day germination of 'Expo Wine' impatiens. The individual means are for 150 seeds.

Priming Time (h)	MTG (h)	Fresh Weight (mg)
0	67.9 b†	402 a
1	67.3 b	389 a
4	64.2 c	405 a
8	65.4 c	355 b
24	76.6 a	323 c
36	76.7 a	320 c
42	76.7 a	284 d

† Means followed by the different lowercase letter are significant different at the P<0.05 level of probability (Student's t test).

Both in control seeds (non-primed) and hydroprimed seeds (in each of the hydropriming times, 1-42 h), the larger the size of the seeds, the less the time to germinate (tables 1 and 4). These results indicate that larger 'Expo Wine' impatiens seeds respond better to hydropriming than smaller seeds. The germination results of non-primed seeds showed that smaller seeds germinated slower than larger seeds (tables 1 and 4), although they achieved 100% germination by 7d after planting (figure 1). These findings demonstrate that the initial (non-primed) seed vigor of the smaller seeds may have been lower than larger seeds. Since seed vigor is a factor influencing seed priming response (Perkins-Weazie and Cantliffe, 1984), it is possible that the lower vigor found in the smaller seeds resulted in less response to hydropriming than larger seeds.

Table 4. Seed hydropriming treatments (0, 1, 4, 8, 24, 36, or 42 h) for six different seed size classes in relation to the mean time to germination (MTG), and seedling fresh weight (50 seedlings) on the 7th day germination of 'Expo Wine' impatiens. The individual means are from 150 seeds. **A** indicates seed diameter > 1.5mm; **B** indicates seed diameter between 1.4 and 1.5mm; **C** indicates seed diameter between 1.3 and 1.4 mm; **D** indicates seed diameter between 1.2 and 1.3 mm; **E** indicates seed diameter between 1.1 and 1.2 mm, **F** indicates seed diameter < 1.1mm.

Seed Size	MTG (h)	Fresh Weight (mg)
A	62.4 e†	454 a
B	67.3 d	408 b
C	69.0 c	382 c
D	72.1 b	326 d
E	73.6 b	294 e
F	76.9 a	262 f

† Means followed by the different lowercase letter are significant different at the P<0.05 level of probability (Student's t test).

Since smaller seeds have a higher surface area/volume ratio compared to larger seeds, they are normally hydrated more rapidly than larger seeds. This conclusion has been reported by Muchena and Grogan (1977) for maize (*Zea mays* L.) and was also found for impatiens seeds in this study (figure 2). After hydropriming for the same period, smaller seeds achieved fuller hydration before larger seeds. As a result, these different levels of hydration cause different responses to hydropriming. Further, excessive hydration found in the smaller seeds might also cause desiccation injury following dry-down after the hydropriming treatment resulting in poorer performance (tables 1 and 4).

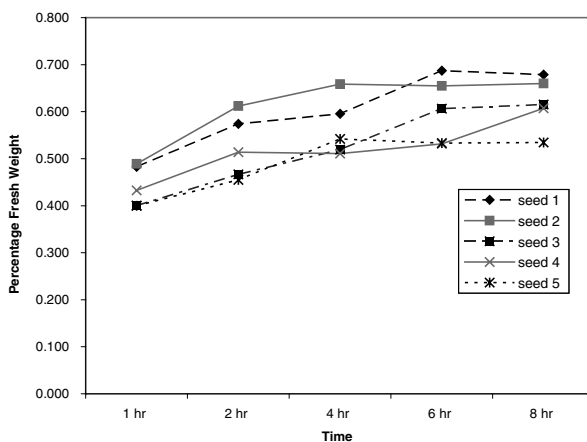


Figure 2. Percentage increase in seed fresh weight for five different size classes (see M&M) of ‘Expo Wine’ impatiens seeds for 1, 2, 4, 6, and 8 hours hydropriming.

Based on these results that show larger impatiens seeds respond better than smaller seeds to hydropriming, successful hydropriming of Impatiens ‘Expo Wine’ seed lots with variations in seed size should be accomplished by screening the seeds into discrete seed size classes, then collecting the larger seeds and conducting the priming treatment. From the MTG results (table 1), ‘Expo Wine’ impatiens seeds, which have 100% TZ viability, do not respond favorably to longer periods of hydropriming (e.g., 24, 36 or 42 h). When primed for shorter durations (e.g., 4 or 8 h), the two larger seed size classes showed faster germination than non-primed seeds; but the smaller seed size classes were not promoted, even after 1 h hydropriming. The longer period of hydropriming might cause seeds to be overprimed at which time irreversible embryo damage may occur during dehydration. It is important to note that although the slow drying method (seeds over saturated salt solutions) was used, the dehydration process may still cause embryo damage. Slow dehydration can only reduce, but not totally eliminate, damage caused during the drying process.

Pandey (1989) reported that priming non-aged French bean (*Phaseolus vulgaris* L. cultivar ‘selection-9’) seeds was deleterious to seed vigor. Priming decreased seedling vigor as manifested by a reduction in seedling fresh weight. He found priming improved vigor in seeds aged for one, two and three years. The seedling vigor increase due to priming was greatest in seeds aged for two years and the greatest reduction in solute leakage due to priming was found in seeds aged for three years.

The reduction in solute leakage after priming indicates that priming increased the membrane integrity of aged seeds. Pandey (1988) postulated the existence of a critical state of seed deterioration at which priming has the maximum potential for relieving ageing predisposed membrane integrity disruption, beyond which it declines and ceases completely in seeds that have lost germinability. Other investigations (Tilden and West, 1985; Pandey, 1988) provide experimental evidence to support the probable existence of metabolic repair, which might facilitate the reversal or alleviation of ageing.

From the above results, it is postulated that when priming non-aged impatiens seeds, there is a critical stage when metabolic repair mechanisms alleviate the damage caused by hydration and dehydration during the priming process. Beyond this stage, repair mechanisms cease and extending the priming time can be deleterious to seed quality as manifested by a reduction in seedling fresh weight (tables 2 and 3). It can also be postulated that non-aged impatiens seeds require less time to arrive at this stage than aged seeds.

Conclusions

Our results demonstrate that a seed size effect in impatiens exists for hydropriming: larger seeds germinate faster than smaller seeds after hydropriming. The larger seeds presumably have greater quantities of stored reserves available for seed germination and may be higher in vigor. Alternatively, smaller seeds may be overprimed. Germination results of primed 'Expo Wine' impatiens seeds with 100% viability indicated that hydropriming for shorter periods (e.g., 4 or 8 h) improved seed vigor, but longer periods (e.g., 24, 36, or 42 h) of hydropriming decreased seed vigor. The optimum priming stage for impatiens 'Expo Wine' occurred within 4 to 8h after hydropriming and beyond that period, deleterious effects associated with desiccation tolerance during dry down caused poorer seed performance.

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