Osmotic priming of true potato seed: Effects of seed age

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Summary

The effects on germination and early seedling growth of presowing true potato seed in water or gibberellic acid (GA) at 1500 ppm and of priming in -1.0, -1.25 and -1.5 MPa solutions of KNO₃ + K₃PO₄ were studied using 30, 18, 6 and 3/4 month-old seed. The influence of light during presowing on the effectiveness of treatments was also investigated.

Overall, priming in the light at -1.0 MPa was the most, and GA the least successful treatment for enhancing emergence and subsequent seedling growth. Though GA increased final emergence from about 20 to 70 % in the most recently harvested lot (3/4 mo), the rate and extent of final germination or emergence in this dormant seed was still much lower than that of the nondormant lots (6-30 mo), especially when the latter were primed. For all lots, dry weight per seedling was 40 % lower in dormant than in nondormant seed, and 20 % higher when seeds were primed at -1.0 MPa than when GA treated. In conclusion, the use of nondormant seed may be a requirement for both effective priming and sowing of potato crops via true seed.

Introduction

The high cost of producing or importing quality seed tubers in developing countries (Horton, 1987) is often a major factor limiting production and consumption of potatoes. Therefore, the International Potato Center (CIP) has given priority to research on innovative alternative methods of potato propagation (Sawyer, 1984). Research on the use of true potato seed (TPS) was started at CIP in 1977 (Mendoza, 1984). More than 40 developing countries are now collaborating with CIP in TPS research, about half of them at farm level; eight are producing potatoes commercially from TPS (Malagamba, pers. commun.).

Most developing countries are in the tropics. TPS technology has potential for increasing potato production in warm-climate areas where seed tuber programmes are hindered by the presence of virus-transmitting aphids. Temperatures higher than 25 °C markedly decrease TPS germination (Clarke & Stevenson, 1943), irrespective of gibberellic acid (GA) presowing treatment (Lam, 1968). Moreover, GA was shown to be ineffective in enhancing emergence of mature seed, i.e. TPS harvested 11 weeks postpollination (Pallais et al., 1987a). Therefore, the use of true seed for warm-climate potato production requires alternatives to GA to circumvent the slow and erratic germination of TPS under supra-optimal temperatures.

In climates where growing seasons are short and high temperatures are common, fast and uniform field emergence is particularly important to crop development. Vigorous early seedling growth decreases the limitations caused by soil crusting and weed

and pathogen infestations. Presowing treatments which improve seed performance under field conditions may prove to be of value for increasing the rate of adoption of TPS technology by farmers in developing countries. One such treatment, which has shown potential for enhancing field emergence under unfavorable conditions in many small-seeded crops, is osmotic priming or osmoconditioning (Brocklehurst & Dearman, 1983; Guedes & Cantliffe, 1980; Haigh et al., 1986; Pill, 1986; Rivas et al., 1984; Sachs, 1977; Szafirowska et al., 1981; Valdes et al., 1985; Wolfe & Sims, 1982). This technique consists of immersing seeds in aerated solutions of an osmotic strength sufficient to prevent radicle emergence while still permitting pregerminative metabolic activity to proceed (Bradford, 1986).

Sadik (1979), working at CIP in Lima, Peru, reported that priming TPS in KNO₃ $+ K_2 PO_4$ at -12.5 bars reduced the time required for germination. A subsequent attempt to apply this technique, however, resulted in a very low percent of germination (Bryan, pers. comm.). Conflicting reports about the effects of priming in some crop species have been attributed to differences in quality or vigor of the seed lots, rather than to varietal characteristics (Brocklehurst & Dearman, 1983; Szafirowska et al., 1981). For priming to be effective in lettuce, high-quality seed must be used (Perkins-Veazie & Cantliffe, 1984). Ells (1963) and Malnassy (1971) found no differences in the effect of inorganic and organic osmotica on the performance of selected vegetable seeds; Sachs (1977), however, did observe differences between them, as well as among different inorganic osmotica. The effectiveness of priming has also been shown to be strongly dependent on the duration of treatment and on the temperature and water potential of the priming solution (Heydecker, 1978). Additional influences include light and aeration conditions during priming (Guedes & Cantliffe, 1980). Though the possible influence of seed age on the response to priming has not been studied, it deserves especial attention in the case of TPS, since this seed is dormant at harvest (Simmonds, 1963).

The objectives of the present study were: 1) to investigate the effects of priming TPS with $KNO_3 + K_3PO_4$ on the rate and extent of in vitro germination and in vivo emergence and dry matter production of young seedlings, and 2) to determine the influences of seed age and light on the effectiveness of priming.

Materials and methods

Seed lots. The TPS lots (S) used in this study were produced during 1984, 1985, 1986, and 1987 using field-grown mother plants of cv. Atzimba crossed with clone R128.6 (cross A). Seed lots were extracted from potato berries borne on hand-emasculated and pollinated flowers. Unless otherwise stated, the seed was produced in Lima, Peru (altitude 50 m, latitude 12° South). To ensure high quality of the seed, supplemental nitrogen (N) was applied during seed development (Pallais et al., 1987b) and optimum maturity was allowed before harvest (Pallais et al., 1987a). Two different lots (I, II) were produced in 1986. The '86-I lot was untreated (no N); '86-II was produced by mother plants which received, starting at emergence, 540 kg/ha of N divided in 12 equal weekly applications. The TPS was stored dry (4-6% moisture content) at 22 °C for about 30, 18, 6, and 3/4 months after harvest ('84, '85, '86, and '87 seed lots, respectively). Two experiments were conducted at CIP's La Molina Experiment Station in Lima, Peru from February to April, 1987. A summary description of materials and methods is presented in Table 1.

Factorial design	Experiment 1	Experiment 2
Seed lots (S)		
'84	X	X
'85	X	X*
'86-I	X	Х
'86-II	х	Х
'87		X**
Presowing treatments (T)		
Priming at -1.0 MPa	х	Х
– 1.25 MPa	Х	
– 1.5 MPa	х	Х
Soaking in GA at 1500 ppm	х	х
Soaking in water	x	х
None (untreated/dry)	X	
Light conditions (L)	Dark	Dark and light
Testing Temperature	Seedling 30 °C (±3°)	Germination and seedling 30/25 °C 28 °C (±5°)

Table 1. Summary of materials and methods.

* The '85 seed from Lima was insufficient; a substitution was made using an '85 lot produced and stored under unknown conditions in Huancayo (3300 m), Peru.

** Produced in Osorno, Chile (41 °SL), April 1987.

Presowing treatments. In the first experiment, presowing treatments (T) were conducted under continuous darkness (dark), and in the second experiment under both dark and alternating 12 h light and dark periods (light). Four samples of 100 (for the germination test) and 25 (for the emergence test) seeds per treatment were obtained at random by the pie method of sampling as described by Ellis et al. (1985). A Dew Point Microvoltmeter (Wescor HR-33T) was used to measure the water potential of the priming solutions. Seeds were evenly spaced inside covered 9 cm Petri dishes lined with Whatman No. 1 filter paper moistened with 5 ml of solution or left dry (untreated). The Petri dishes were sealed with Parafilm M and kept at 15 °C. Seeds were primed or untreated dry for five days. GA and water treatments were started on the fourth day. After treatments, all seed lots were rinsed with running water for at least 2 min and dried over filter paper for 24 h in an incubator, at 15 % relative humidity and 20 °C. They were then sealed over fresh silica gel and stored at room temperature (about 22 °C) for five days before testing.

Seed testing. In the germination test, seeds were evenly distributed in Petri dishes over moist filter paper; water was added as needed. The germination environment was at a temperature of 30/25 °C under 12 h alternating light/dark conditions for the first 24 h and thereafter under continuous darkness; seeds were also briefly exposed to light in the laboratory during watering and daily evaluations which lasted less than 30 min. In the seedling tests, seeds were sown inside polystyrene flats in a steam sterilized soil mixture of 1 peat moss:1 sand, at 1 cm deep and with 1.5 cm between plants and 5 cm

between rows at field capacity. Seeds were covered with soil immediately after sowing, or after 24 hours in the second experiment. Seedling tests were conducted in a water-cooled glasshouse and temperature was monitored continuously with a hydrothermograph. The first appearance of a radicle or hypocotyl-hook signified germination or emergence, respectively. Counts were made every 24 h for ten days, commencing the day after seeding or sowing. Factorial statistical analyses were based on the coefficient of velocity (CoV) of germination or emergence 1-10 days after sowing (Scott et al., 1984), on final percent emergence after 17 days, and on dry weight per seedling (tops).

Results

Experiment 1. For all lots, priming at -1.0, -1.25, or -1.5 MPa resulted in a significantly higher rate (CoV) of seedling emergence than nonpriming (GA, water, and dry) treatments (Table 2). Priming at -1.0 was the best treatment. GA-treated seed had the lowest CoV value. For all treatments, the '84 seed had the highest CoV, and '86-I the lowest. Mean final emergence was also significantly higher (>96 %) when primed at either -1.0 or -1.25, but there was no difference between priming at -1.5 and non-priming treatments. Final emergence was highest in the '85 lot (96 %). The '84 lot was intermediate (93 %), and there was no difference between the two '86 lots (89 %).

Experiment 2. Interaction effects of $S \times T$, $S \times L$, and $T \times L$ (see Table 1 for explanation of symbols) were significant on the resulting CoV's in both germination and emergence tests (Figs. 1a-c), though the $S \times T \times L$ interaction was not significant (P < 0.05) in either test. Since only less than 2 % of the '87 seed germinated irrespective of treatments, these data are not presented nor are they included for the analysis of the results

TPS lot	Emergence rate (CoV)						Final percent emergence							
	priming (– MPa)			nonpriming			priming (–MPa)			nonpriming				
	1.0	1.25	1.5	GA	H ₂ O	Dry	Mean	1.0	1.25	1.5	GA	H ₂ O	Dry	Mean
'84	17.9	16.9	17.4	15.3	15.5	15.7	16.4a	98	90	91	92	92	93	93b
'85	18.1	17.5	15.6	12.3	15.0	15.9	15.7b	100	99	99	97	92	90	96a
'86-I	16.8	16.3	14.8	13.5	14.7	14.3	15.1c	96	98	98	84	89	89	89c
'86-II	16.3	16.1	16.6	14.0	14.6	15.0	15.4bc	94	97	89	90	82	82	89c
Mean	17.3a	16.7b	16.1c	13.8e	15.0d	15.2d		97a	96a	90b	91 b	89b	89b	
Signi	ficance													
TPS lot (S) < 0.001)1			0.013								
Treatm. (T) < 0.001					0.011									
$S \times T$	S×T 0.055					0.434								
cv (%	0)			6.9							9.1			

Table 2. Effect of presowing treatments on the coefficient of velocity (CoV) of seedling emergence and final percent of emergence in four different TPS lots of cross A.

Means followed by the same letters are not significantly different at P < 0.05.

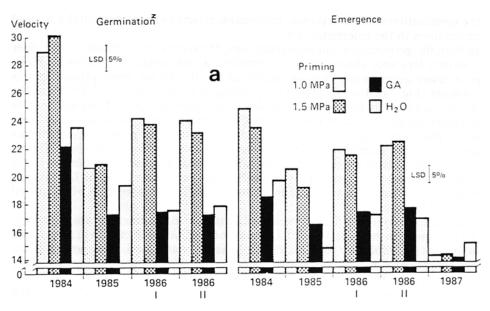
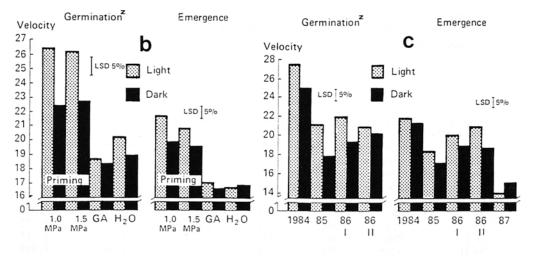


Fig. 1a. Effect of presowing treatments on the coefficient of velocity of germination and emergence in five TPS lots of cross A.



²1987 failed to germinate.

Fig. 1b. Effect of presowing treatments on the coefficient of velocity in five TPS lots. Fig. 1c. Influence of light on the effect of presowing treatments.

in the germination test. Nevertheless, measurable effects of treatments on the '87 seed were obtained in the emergence test.

In both the germination and emergence tests, the effects of treatments in the older (6-30 mo) lots were similar to those obtained in the previous experiment. However, a faster germination rate was exhibited by the '84 lot when primed in this experiment (Fig. 1a). For the older lots, priming at either -1.0 or -1.5 MPa in the light was significantly more effective in enhancing germination and emergence rates than nonpriming (Figs. 1a, c). However, the CoV values of the '87 seed, markedly lower than in the older lots, were not significantly affected by presowing treatments (Figs. 1a, c). In the germination test, a CoV response to treatments somewhat different than in the previous experiment was found in the '85 Huancayo seed. Priming (at either osmotic level) was not significantly different from presowing in water for this lot, though the CoV values in all treatments were significantly higher than when treated with GA (Fig. 1a).

Priming in the light significantly increased germination and emergence rates as compared to priming in the dark (Fig. 1b). Light did not similarly affect rates in nonprimed seed, though it did enhance germination rate in water-treated seed. Light generally had an enhancing effect on most lots, with the exception of '87 seed in which the emergence rate was significantly higher when presown in the dark (Fig. 1c).

Differing responses to presowing treatments between the older lots and the '87 seed were also observed with regard to final emergence (Table 3). Emergence was above 91 % for the older lots, except for '85 Huancayo seed when treated with water (82 %). In contrast, seed emergence in the '87 lot was 69 % when treated with GA and 24 % or less for all the other treatments.

All three experimental factors (S, T, L) significantly affected the resultant dry weight per seedling and, as Table 4 shows, there were no significant interactions among these factors. The mean dry weight of seedling tops produced by the older lots was at least 40 % greater than that of the '87 seedlings. For all lots, seedling dry weight when primed at either -1.0 or -1.5 MPa was higher than when nonprimed. The data also indicated that presowing in the light increased dry weight over presowing in the dark.

TPS lots	Priming	(–MPa)	Nonpri	ming	Signific.	Р	
	1.0	1.5	GA	H ₂ O			
'84	98a	96a	92a	96a	S×T	< 0.001	
'85	98a	93a	98a	82b	S×L	0.772	
'86-I	98a	96a	99a	96a	T×L	0.115	
'86-II	92ab	98a	97a	94a	S×T×L	0.099	
'87	21d	24d	69c	8e	cv (%)	8.4	

Table 3. Effect on final emergence of presowing treatments (T) under light or in the dark (L) in five TPS lots (S) of cross A.

Means followed by the same letters are not significantly different within and between columns at P < 0.001.

TPS lot		Presowing treatments		Light		Signific.	Р
'84 '85 '86-I '86-I1 '87	3.37a 2.98a 3.12a 3.23a 1.84b	Priming – 1.0 MPa Priming – 1.5 MPa GA 1500 pm Control H ₂ O	3.25a 3.05a 2.63b 2.70b	Light Dark	3.05a 2.77b	S×T S×L T×L S×T×L cv (%)	0.455 0.645 0.157 0.878 29.0

Table 4. Mean dry weight per seedling 17 days after sowing in five TPS lots of cross A in which various presowing treatments were applied under light and dark conditions.

Means followed by the same letter are not significantly different at P < 0.05.

Discussion

This study demonstrated that priming of TPS in $KNO_3 + K_3PO_4$ was more effective as a seed presowing technique for optimizing germination and seedling performance than the standard GA and control treatments. However, in treatments applied less than one month after the seed was harvested ('87 lot), final emergence was about 50 % higher when presown with GA than when primed (Table 2). Therefore, the '87 seed lot can be considered to have been dormant at the time of testing (3/4 mo). This indicates that the use of nondormant TPS is a requirement for effective priming. The lack of response of TPS to priming previously observed at CIP (Bryan, pers. commun.) could have been due to the dormancy state of the seed.

Priming at -1.0 MPa was generally more effective than at -1.5. The effectiveness of priming was increased by light; light during presowing treatments enhanced germination and emergence rate in most lots (Fig. 1c). However, presowing of dormant TPS in the light lowered emergence rate (Fig. 1c) in the few (< 8 %) '87 seedlings that emerged during the first ten days following sowing. Sensitivity to light for seed germination during storage, coupled with complex temperature interactions, has been well documented for lettuce (*Lactuca sativa*) seed of the Grand Rapids variety (Mayer & Poljakoff-Mayber, 1975). There are conflicting reports as to the effect of light on germination of potato seeds. Simmonds (1963) demonstrated that TPS germination was stimulated by light; Lam & Erickson (1966) concluded that germination of recently harvested seed was greater in darkness. Although light treatment was applied during presowing in this study and not during germination, the data (Fig. 1c) may help to explain the differences between these findings.

The widespread belief that GA effectively breaks TPS dormancy was not supported by the results of lower percent of emergence in GA-treated dormant seed as compared to nondormant, irrespective of presowing treatment (Table 3). The inadequacy of GA to fully induce germination in fresh TPS was also recently reported by D'Antonio & McHale (1988). An additional indication that GA is not a solution for releasing newly-harvested TPS from dormancy effects was the 40 % lower seedling dry matter production in dormant seed versus nondormant. These results support the conclusions of Taylorson & Hendricks (1977) that although light-requiring seeds can be stimulated to germinate by GA, no seeds seem to be solely dependent on exogenous GA to be totally released from dormancy. These findings also support the suggestion previously

made (Pallais, 1987) that the use of GA to break TPS dormancy must receive closer attention. Moreover, even the appropriateness of this hormone in clonal breeding programmes may be questioned by these results. For example, what if the response to GA in dormant TPS is genetically controlled and the 31 % of the seed which was not induced to emerge by GA contains important gene combinations?

In conclusion, the performance of nondormant TPS during germination, emergence, and early seedling growth was best enhanced by priming in $KNO_3 + K_3PO_4$ at -1.0 MPa for 5 days at 15 °C in the light. Dormant TPS did not respond to priming; this seed may require an after-ripening period of at least six months for effective priming before sowing. After-ripening under dry storage conditions results in increased ability to germinate in seeds of many species (Mayer & Poljakoff-Mayber, 1975). The specific length and conditions of storage to best satisfy this requirement for TPS deserve to be investigated. However, we must recognize that the significance of physiological enhancement (e.g. priming and after-ripening) may not compare to the improvements which are likely to result from breeding and selection for more vigorous (during early seedling development) and less dormant TPS crosses. The fact that genetic variability of germination characteristics exists in cultivated potatoes has been demonstrated by Simmonds (1964), and more recently by Pallais et al. (1987c). Moreover, studies of a few natural populations of weeds indicate that seed dormancy may be readily selected out of a population (Thompson, 1981; Witcombe & Whittington, 1972). Finally, it is proposed that if and when the character of seed dormancy is selected out of TPS crosses, seed priming may still be a useful technique for optimizing true seed vigor in potatoes.

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