Integrating Low Water Potential Seed Hydration with Other Treatments to Improve Cold Tolerance

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Matricconditioning improved the performance of pepper, tomato, sweet corn, snap bean, table beet, sugar beet and watermelon seeds in early field plantings at suboptimal temperatures (averaged over 10 d after planting) ranging from 12 to 18 °C. Reduction in the time to 50% (T_{50}) emergence in conditioned seeds ranged from 0.6 d in watermelon to 3.3 d in pepper and improvement in emergence from 10% in sugar beet to 30% in table beet. Further improvement in emergence occurred by inclusion of pesticides and/or gibberellin during conditioning. A 4 d conditioning of pepper at 25 °C was superior to 7 d conditioning at 15 °C in seeds germinated at 15 °C on filter paper, but 15 °C conditioning was superior in improving percentage emergence in early field plantings. Tomato seeds conditioned at 15 or 25 °C performed equally well in the field. A 2 d conditioning was superior to 1 d conditioning in improving the performance of supersweet sweet corn cultivars grown in a growth chamber at 10/20 °C. The water uptake rate in the presence of Micro-Cel E during matricconditioning of sweet corn seeds was slower than when the seeds were exposed to the same amount of water in absence of the carrier. Electrolyte leakage was greater in supersweet 'Challenger' sweet corn seeds carrying the *sh₂* gene compared to the *sugary* type sweet corn 'More', and in both cases matricconditioning reduced the leakage. Lettuce seeds matricconditioned for 24 h had higher 1-aminocyclopropane-1-carboxylic acid (ACC) content, developed greater ACC oxidase activity and performed better at 10 °C (germinated earlier and had higher percentage germination) than the untreated seeds. Matricconditioning appears to bring about beneficial physical, physiological and biochemical changes that seemingly improve embryo growth potential and tolerance to low temperatures.

Key words: Seed matricconditioning, seed osmoconditioning, imbibitional chilling injury, water uptake, solute leakage, vegetable seeds.

INTRODUCTION

Imbibitional chilling injury is a common feature affecting many seeds but is particularly severe on large seeds such as soybeans, snap beans and sweet corn and seeds of warm season crops, such as tomato, pepper and watermelon. Even seeds that are usually regarded as tolerant of low temperature, such as table beet, carrot, and onion, germinate poorly and have poor stand (final number of plants) when exposed to low temperature.

Imbibitional injury is related to rapid uptake of water by the seed and is often made worse by low temperatures. Several approaches have been used to decrease the impact of low temperature on the performance of seeds. These include increasing seed hydration levels prior to planting (Bennett and Waters, 1987), using seeds with semi-hard seed coats to slow the water absorption rates (Taylor and Dickson, 1987), coating seeds with hydrophobic substances (Priestley and Leopold, 1986), and subjecting them to low water potential (ψ) seed hydration (Liu, Chen and Zheng, 1991; Khan, 1992).

Presowing treatment of seed in low ψ solutions of

polyethylene glycol (PEG) and salts has been shown to improve the performance of a number of seeds at suboptimal temperatures in laboratory and/or field plantings (see Khan, 1992). In addition to osmotic treatment of liquid media via dissolved solutes, matric forces of solid carriers devoid of osmotic solutes (such as vermiculite, calcined clay and Micro-Cel E) have been utilized to control hydration of seeds (Bennett and Waters, 1987; Kubik *et al.*, 1988; Khan *et al.*, 1990; Parera and Cantliffe, 1991). Such terms as moisturizing (Bennett and Waters, 1987), solid matrix priming (Kubik *et al.*, 1988; Eastin, 1990) and matricconditioning (Khan *et al.*, 1990) have been used to describe the presowing seed conditioning with moist solid carriers with matric properties. Improvements in the rate of germination and germination percentage by osmoconditioning or matricconditioning have generally been observed at suboptimal temperatures (Khan, 1992).

This paper describes the effect of seed matricconditioning on improving performance of seeds at suboptimal temperatures in a field setting and some of the physical and biochemical changes associated with matricconditioning. These results are discussed in light of current understanding of how low ψ seed hydration lessens imbibitional chilling injury.

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MATERIALS AND METHODS

Seeds

Following seeds were used: lettuce (*Lactuca sativa* L. cv. Mesa 659), sweet corn (*Zea mays* L. cvs. More, Mariah, Phenomenal, GSS 3492, Showcase, Supersweet Jubilee and Challenger), snap bean (*Phaseolus vulgaris* L. cv. Labrador), table beet (*Beta vulgaris* L. cv. Ruby Queen), sugar beet (*Beta vulgaris* L. cv. E-4), tomato (*Lycopersicon esculentum* Mill. cv. Heinz and Super Marmande), pepper (*Capsicum annuum* L. cv. El Paso), carrot (*Daucus carota* var. *sativus* cv. Long Emperor) and watermelon [*Citrullus lunatus* (Thumb.) Matsumi & Nakai cv. Tri-X triploid]. The seeds were kept at 7 °C and 28% relative humidity (RH) in nonsealed containers and were equilibrated with ambient atmosphere for at least 2 d before use. The initial moisture contents of bean and sweet corn seeds were 13 and 11%, respectively, while those of others ranged from 5 to 7%.

Matricconditioning

Matricconditioning procedure has been described in detail elsewhere (Khan *et al.*, 1992). It consists of mixing the seed with water and Micro-Cel E, a synthetic calcium silicate, with high water holding capacity. The ratios of seed to Micro-Cel E to water in the conditioning mixtures, the duration and temperature of conditioning and the ψ of Micro-Cel E were as follows: sweet corn, 1:0.2:1.4 (1 or 2 d conditioning at 15 °C, $\psi = -0.5$ MPa); snap bean, 1:0.4:1.3 (2 d conditioning at 15 °C, $\psi = -1.4$ MPa); table beet, 1:0.2:1.15 (7 d conditioning at 15 °C, $\psi = -0.5$ MPa); sugar beet, 1:0.25:0.63 (7 d conditioning at 15 °C, $\psi = -1.4$ MPa); tomato and pepper, 1:0.3:1.38 (7 d conditioning at 15 °C, $\psi = -1.0$ MPa) and 1:0.3:1 (4 d conditioning at 25 °C, $\psi = -1.2$ MPa); watermelon, 1:0.25:1 (4 d conditioning at 25 °C, $\psi = -1.0$ MPa); lettuce, 1:0.25:1.5 (1 d conditioning at 15 °C, $\psi = -0.7$ MPa). Carrier ψ potential (= matric potential) was estimated by first determining the water content of Micro-Cel E at equilibrium with the seed at the end of conditioning and then relating the water content to the matric potential (Khan *et al.*, 1992). Carrot, tomato and pepper seeds were dried by forced air at 25 °C for 2 h without removing Micro-Cel E, the loose carrier that came off the seeds was removed by sieving. Sweet corn seed was dried for 24 h (in the leakage study, seeds were used without drying) and the carrier that came off was removed by sieving. Table beet, sugar beet and lettuce seeds were rinsed before drying for 2 h. Snap bean seeds were sieved to separate them from Micro-Cel E and were planted without drying.

Pesticides (fungicides and insecticide), gibberellin (GA₃) and biocontrol agent (AB254 strain of *Pseudomonas aureofaciens* kindly provided by Dr Nancy Callan) were applied to seeds either via methylcellulose (an adhesive) or were suspended or solubilized in water used for conditioning. Apron (metalaxyl) and thiram were applied to table beet, sugar beet and watermelon seeds as a suspension in water used for conditioning at the rate of 50 and 200 mg active ingredient per 160 g seeds. Lorsban [an insecticide, *O,O*-diethyl *O*-(3,5,6-trichloro-2-pyridinyl) phosphothionate],

Apron and demosan were applied to snap bean seeds via 1.5% methylcellulose slurry at the rate of 100, 40 and 260 mg active ingredient per 160 g seeds and the bean seeds were dried overnight before conditioning. GA₃ at 2 μ M was added to water at the time of conditioning. Sweet corn seeds were commercially treated with Apron, thiram, captan, imazalil and Vitavax (carboxin). The amounts of pesticides applied to various nonconditioned seeds were identical to those applied to conditioned seeds, except that the pesticides were applied via 1.5% methylcellulose and the seeds were dried overnight. Bacterial treatment of sweet corn consisted of scraping an actively growing culture of *Pseudomonas aureofaciens* from a 9 cm plate and suspending it in water used for conditioning.

Germination and emergence

Pepper and lettuce seeds, in three replicates of 50 seeds each, were germinated at 15 °C and 10 °C in 9 and 5 cm Petri plates, respectively, lined with two layers of Whatman No. 1 filter paper moistened with 6 and 3 ml of water. Germination (radicle protrusion) was scored on a daily basis. Twelve sweet corn seeds per row per plastic box (35 × 26 × 11 cm) in three replications were planted 1 cm deep in a 1 peat:1 vermiculite mix, and seedling emergence was determined in a growth chamber set at 20 °C for 12 h in light and at 10 °C in darkness. Shoot fresh weight was determined after 14 d. In field trials, the effects of treatments were evaluated by planting seeds in Lima silt loam (fine-loamy, mixed, mesic Glossoboric Hapludolf). Seeds were planted in 6-m rows, 75 cm apart with 100 seeds per row at a depth of 2.5 cm (tomato, pepper, carrot, table beet and sugar beet) and 5 cm (snap bean, sweet corn and watermelon) using a cone or a flex seeder. Treatments were replicated six times in a randomized complete block design. Seedling emergence was scored at 1- to 2-d intervals after planting. Time to 50% (T₅₀) emergence was computed based on final percentage emergence. The maximum and minimum daily temperatures at a 5-cm depth were recorded (temperature data at 2.5 cm depth were not available) and temperatures averaging over first 10 d after planting of each crop were computed.

Water uptake and electrolyte leakage

Water taken up by Challenger sweet corn seeds in presence of 4 g Micro-Cel E (matricconditioning mixture: 20 g seeds, 4 g Micro-Cel E and 28 g water) and in its absence was determined in a 0.5-l glass jar at 15 °C. The initial and final moisture content of the seeds were determined by the air-oven method as described previously (Ptasznik and Khan, 1993). Logarithmic equations were generated to fit the relationship of water content to the soak duration in presence and absence of Micro-Cel E. To determine electrical conductivity (E.C.) in the seed leachate, untreated and matricconditioned seeds were rinsed for a few seconds with deionized water. Twenty-five seeds were soaked in 50 ml deionized water for 7 h at 25 °C in light. Levels of electrolyte leached during soaking were determined by a digital conductivity meter (VWR, Rochester, NY, USA) and E.C. was expressed in μ S cm⁻¹ g⁻¹ seeds.

TABLE 1. Improved performance of matriconditioned seeds at low soil temperatures in early field plantings at Geneva, New York, USA

Seed/cultivar	Treatment	Soil temperature§ (°C)	Planted	T ₅₀ † (d)	% Emergence
Sweet corn	Noncond (F)†	13.5	14 May 1993	13.8 ^{a*}	50 ^b
(Supersweet Jubilee)	Cond (F)	13.5	14 May 1993	12.5 ^b	60 ^a
Snap bean	Noncond (L)	12.2	3 May 1991	13.4 ^a	38 ^c
(Labrador)	Cond (L)	12.2	3 May 1991	12.5 ^b	58 ^b
	Cond (A + D + L + GA)	12.2	3 May 1991	12.6 ^b	92 ^a
Table beet	Noncond	12.0	1 May 1990	18.3 ^a	70 ^c
(Ruby Queen)	Noncond (A + T)	12.0	1 May 1990	16.5 ^b	100 ^b
	Cond	12.0	1 May 1990	14.5 ^c	100 ^b
	Cond (A + T)	12.0	1 May 1990	14.8 ^c	119 ^a
Sugar beet	Noncond (A + T)	16.2	1 June 1990	10.0 ^a	45 ^b
(E-4)	Cond (A + T)	16.2	1 June 1990	8.0 ^b	55 ^a
Tomato	Noncond	12.8	8 May 1992	9.9 ^a	45 ^b
(Heinz)	Cond	12.8	8 May 1992	6.9 ^b	71 ^a
Tomato	Noncond	12.8	8 May 1992	9.0 ^a	46 ^b
(Super Marmande)	Cond	12.8	8 May 1992	6.5 ^b	70 ^a
Pepper	Noncond	13.4	13 May 1992	19.3 ^a	43 ^b
(El Paso)	Cond	13.4	13 May 1992	16.0 ^b	60 ^a
Carrot	Noncond	12.4	2 May 1992	17.8 ^a	37 ^b
(Long Imperator)	Cond	12.4	2 May 1992	15.1 ^b	51 ^a
Watermelon	Noncond (A + T)	18.2	10 June 1992	6.4 ^a	50 ^b
(Tri-X 313 triploid)	Cond (A + T)	18.2	10 June 1992	5.0 ^b	80 ^a

* Mean separation in each seed type by l.s.d. ($P = 0.05$).

† Chemicals used during conditioning: L, Lorsban; A, Apron; T, thiram; F, combination of captan, thiram, Apron, imazalil and Vitavax. GA, GA₃.

‡ Time for 50% of final emergence percentage.

§ Daily soil temperatures (maximum and minimum) averaged over first 10 d after planting.

ACC content and ACC oxidase activity

The contents of 1-aminocyclopropane-1-carboxylic acid (ACC) in untreated and 24 h matriconditioned (seeds were dried back to the level of untreated seeds) Mesa 659 lettuce seeds were determined as described previously (Huang and Khan, 1992; Khan and Andreoli, 1993). Briefly, seeds (in three replications) were homogenized with 80% methanol using a mortar and pestle. The homogenate was centrifuged at 5000 g for 20 min and the supernatant dried *in vacuo* at 45 °C. The ACC content in the dried extract was determined by chemical degradation of ACC to ethylene according to the method of Lizada and Yang (1979). For ACC oxidase determination, 50 untreated and conditioned seeds (three replications) were placed in 5.7 ml tubes in the presence or absence of 0.2 ml 1 mM ACC, the tube sealed with a rubber septum and incubated for 4 h at 10 °C. After incubation, 1 ml gas sample was withdrawn from the tube and injected into a Hewlett Packard 5890 Series II gas chromatograph equipped with flame ionization detector and a 183 × 0.32 cm stainless steel column containing Poropak Q. The increase in ethylene produced over that of the untreated (–ACC) blank represented a measure of ACC oxidase activity.

RESULTS AND DISCUSSION

Matriconditioning to improve performance of seeds at suboptimal temperatures

Seed matriconditioning, with or without the addition of various chemicals, reduced the T₅₀ of emergence and

improved the final emergence percentage at suboptimal temperatures in early field plantings of a number of vegetable seeds (Table 1). The seeds were planted in May and June when the soil temperature, averaged over the first 10 d after planting, ranged from 12 to 18.2 °C for different vegetables. Depending upon the seeds the reduction in T₅₀ of emergence ranged from 1 to 3 d. Improvement in final emergence percentage by conditioning alone ranged from 10 to 30%. Fungicides and/or hormone added to table beet and snap bean seeds during conditioning had little effect on T₅₀ of emergence when compared to seeds conditioned in absence of the chemicals, but significantly improved the final emergence percentage (92 *vs.* 58% for beans and 119 *vs.* 100% for table beet) (greater than 100% emergence in table beet is due to the fact that a multigerminant cultivar was used).

Effect of matriconditioning tomato and pepper seeds at 15 and 25 °C on performance at low temperatures

Matriconditioning pepper seeds at 15 °C for 7 d or at 25 °C for 4 d reduced the time needed for germination on filter paper at 15 °C and conditioning at 25 °C was more effective than conditioning at 15 °C in reducing the germination time (Fig. 1). When the performance of seeds conditioned at 15 and 25 °C was evaluated in early field plantings on 13 May (soil temperature averaged over 10 d after planting: 13.4 °C) or 10 June 1992 (soil temperature averaged over 10 d after planting: 18.2 °C), seeds conditioned at 15 °C outperformed those conditioned at 25 °C with respect to final emergence percentage (Table 2). These

FIG. 1. Effect of matricconditioning 'El Paso' pepper seeds on germination at 15 °C. Seeds were conditioned for 4 d at 25 °C and for 7 d at 15 °C.

data indicate that adverse changes occur in pepper seeds during 25 °C conditioning, which are expressed under cold wet conditions in the field. For tomato seeds, which have a shorter germination and emergence time than pepper, a 4 d conditioning at 25 °C or 7 d at 15 °C was equally effective in improving the performance of seeds in early field plantings on 8 May 1992 (soil temperature averaged over 10 d after planting: 12.8 °C) (Fig. 2). Tomato seeds germinated and emerged more rapidly than pepper seeds and thus might have escaped the damaging effect of cold temperature and soil pathogens.

Improving performance of supersweet corns at low temperature by matricconditioning

Sweet corns carrying the *shrunk-2* (*sh₂*) mutant gene are referred to as supersweet corns. Several factors contribute to poor stand establishment of these seeds. These include low vigour, susceptibility to imbibitional damage and susceptibility to seedborne and soilborne diseases. Presowing seed conditioning with Micro-Cel E in the presence of appropriate fungicides proved effective in improving biomass production (shoot growth) in several cultivars in a growth chamber study at a suboptimal temperature regime (10/20 °C) (Fig. 3). A 2 d conditioning generally appeared to be superior than 1 d conditioning. Matricconditioning of sweet corn seeds also improved the percentage emergence in a 14 May 1993 planting (soil temperature 13.5 °C, averaged over 10 d after planting) (Fig. 4). Addition of a strain (AB254) of *Pseudomonas aureofaciens* during conditioning had no effect on emergence. Other solid carriers have also been used to condition sweet corn seeds. Conditioning (solid matrix priming) with expanded calcined clay in the presence of fungicides or NaOCl improved the emergence percentage over nonconditioned seeds, but not over those treated with fungicides alone, in field plantings at temperatures ranging from 20 to 30 °C during the first 7 d after planting (Parera and Cantliffe, 1992).

TABLE 2. Effect of matricconditioning 'El Paso' pepper seeds planted in the field on 13 May and 10 June 1992

Treatment	13 May		10 June	
	T ₅₀ (d)*	Emergence (%)†	T ₅₀ (d)*	Emergence (%)†
Untreated	19.4 [§]	43 ^b	11.3 ^a	45 ^b
4 d conditioning at 25 °C	11.2 ^c	39 ^b	7.7 ^b	55 ^{ab}
7 d conditioning at 15 °C	16.0 ^b	60 ^a	8.7 ^b	68 ^a

* Time for 50 % of final emergence percentage, 34 d after planting.

† Emergence 34 d after planting.

§ Mean separation by l.s.d. (*P* = 0.05).

FIG. 2. Effect of matricconditioning 'Super Marmande' tomato seeds on field emergence in a 8 May 1992 planting in Geneva, New York, USA. Seeds were conditioned (COND) for 4 d at 25 °C and for 7 d at 15 °C. Bars adjacent to symbols represent l.s.d. (*P* = 0.05). UNT, Untreated.

Reducing water uptake rate and increasing water content

Water uptake rate during matricconditioning of sweet corn seeds was slower than when seeds were imbibed directly in the same amount of water used for conditioning (Fig. 5). Logarithmic curves generated for the relationship of seed water content and soak duration provided an excellent fit ($r^2 = 0.96$ to 0.98). Seed coating with hydrophobic substances, such as lanolin and polyvinyl alcohol as well as osmoconditioning in PEG have been reported to reduce water uptake and chilling injury in soybean, corn and cotton seeds by providing time for repair and recombination of biomembranes (Priestley and Leopold, 1986; Tao, Zuo and Cheng, 1991; Yang, Chen and Guo, 1992). Semi-hard bean seeds, with an ability to slow the rate of water uptake, had greater tolerance to imbibitional chilling injury than the seeds with more permeable coats (Taylor and Dickson, 1987).

FIG. 3. A, Seedling emergence at 20/10 °C in a peat-vermiculite mix following sowing of matriconditioned (for 0, 1 and 2 d) 'Challenger' supersweet corn seeds. Bars denote mean separation by l.s.d. ($P = 0.05$). B, Effect of matriconditioning seeds of supersweet cultivars for 0, 1 and 2 d on seedling growth as determined by changes in shoot biomass. Shoot weight was determined after 14 d. Mean separation in each cultivar by l.s.d. ($P = 0.05$). UNT, Untreated.

FIG. 4. Effect of 2 d matriconditioning 'Supersweet Jubilee' sweet corn seeds, with (■) or without (□) the addition of a strain AB254 of *Pseudomonas aureofaciens*, on field emergence in a 14 May 1993 planting in Geneva, New York, USA. Controls were: untreated (○); treated only with AB254 (●). Mean separation by l.s.d. ($P = 0.05$).

In addition to reduced water uptake rate, seed water content also influences seed performance at low temperatures. Snap bean seeds with high initial moisture (12%) have been reported to perform better than the seeds with low moisture content (8%) at low temperature (Ladror, Dyck and Silbernagel, 1986), indicating that high moisture

FIG. 5. Rate of water absorption by 'Challenger' supersweet corn seeds at 15 °C in the presence (○) and in absence (●) of Micro-Cel E. Ratios of seed: Micro-Cel E: water in the two cases were: 1:0.2:1.4 (in presence of Micro-Cel E) and 1:0:1.4 (in absence of Micro-Cel E). Logarithmic equations were generated to fit the experimental data. ***, Significant at 0.5% level.

is essential for maintaining the functional integrity of membranes and thus preventing chilling injury. Similarly, germination and emergence of high sugar sweet corn seeds

FIG. 6. Electrolyte leakage from 'More' (a sweet corn carrying the *su* gene) and 'Challenger' (a supersweet corn carrying the *shrunk* gene, *sh₂*) sweet corns, before (▨) and after (■) 24 h matricconditioning at 15 °C. Leakage from conditioned (nondried) and untreated seeds was determined after a 7 h soak at 25 °C. Bars denote l.s.d. ($P = 0.05$).

was improved by a presowing hydration treatment (Bennett and Waters, 1989).

Reduced electrolyte leakage following preconditioning

Supersweet corns are highly susceptible to imbibitional chilling injury and generally perform poorly compared to the more traditional sweet corn cultivars, such as 'More', that carry the *sugary* gene. Electrolyte leakage by supersweet corn 'Challenger' was 2.7 times greater than that by 'More' (Fig. 6). Matricconditioning reduced the electrolyte leakage in 'Challenger' and 'More' by 52 and 35 %, respectively. A similar decrease in electrolyte leakage as a result of matricconditioning was observed in snap bean seeds (Ptasznik, Barnard and Khan, 1992). Using another solid carrier, Parera and Cantliffe (1992) found a decrease in electrolyte leakage following solid matrix priming. Osmoconditioning with liquid carriers has similarly been shown to decrease electrolyte leakage (Pandey, 1989; Weges and Karssen, 1990).

Low temperature adversely influences the functional integrity of biomembranes, nuclei and mitochondria (Yan *et al.*, 1989; Yang *et al.*, 1992). Development of mitochondria from proplastids in plumular root cells was not restored upon transfer of nonconditioned soybean seeds to normal temperature after an exposure to low temperature. However, seeds that were osmoconditioned prior to exposure to low temperature, developed distinct bilayered membrane and tube cristae, indicating that redifferentiation and development of proplastids into mitochondria at low temperature played an important role in the increase of soybean seed vigour and its resistance to chilling injury (Yang *et al.*, 1992).

Enhanced ACC production and utilization in relation to cold tolerance

The level of ACC rose during a 24 h matricconditioning of lettuce seeds from 0.6 nmol g⁻¹ to 1.3 nmol g⁻¹ seeds (Table

TABLE 3. Effect of matricconditioning of lettuce seeds on ACC content and ACC oxidase activity in relation to germination at low temperature

Treatment	ACC (nmol g ⁻¹ seed)	ACC-oxidase* (nl ethylene g ⁻¹ seeds)	Germination (%)†	
			24 h	120 h
Untreated	0.6	5	0	75
Conditioned‡	1.3	20	65	96
l.s.d. ($P = 0.05$)	0.3	4	5	7

* Seeds were incubated for 8 h at 10 °C before enzyme assay.

† Germination was determined after 24 and 120 h soak at 10 °C.

‡ Seeds were matricconditioned for 24 h and then dried back to the moisture level of untreated seeds.

3). The ACC-oxidase activity in untreated and conditioned seeds determined following a 8 h soak at 10 °C were 5 nl ethylene g⁻¹ and 20 nl ethylene g⁻¹ seed, respectively. Germination percentages of untreated and conditioned seeds after a 24 h soak at 10 °C were 0 and 65 % and after a 120 h soak 75 and 96 %. These data suggest that the higher level of ACC and greater activity of ACC oxidase in conditioned seeds compared to the untreated seeds may confer low temperature tolerance to these seeds. Improved embryo growth potential and vigour in osmoconditioned and matricconditioned lettuce seeds have been reported under a variety of stressful conditions, including low temperature (Khan, 1977, 1992; Khan *et al.*, 1993). As in lettuce, conditioned pepper seeds had higher ACC level, greater ACC-oxidase activity and greater embryo growth potential than the nonconditioned seeds (Ilyas and Khan, unpubl. res.). Thus, cold tolerance may be related to enhanced ACC production and its utilization. In other studies, osmoconditioned soybean seeds, unlike nonconditioned seeds, retained the ability to increase soluble protein content and activities of a wide variety of enzymes, including acid and alkaline phosphatases, esterases, aldolase, isocitrate lyase, cytochrome oxidase, malate dehydrogenase, catalase and superoxide dismutase (SOD) following imbibition at low temperature and before germination (Liang, Zhao and Zheng, 1990; Liang *et al.*, 1991).

CONCLUSION

It can be concluded that low ψ seed hydration treatments influence physical, biochemical and physiological events associated with improved performance at low temperatures. An important cause of imbibitional chilling injury to young seedlings may be inhibition of reorganization of biomembrane structures and functions. The low ψ seed hydration treatment appears to satisfy two important criteria for improved performance. First, it reduces, particularly in large, chilling sensitive seeds, the rapid water uptake and the associated injury to the membrane system. Second, it extends the lag phase of imbibition by suspending germination, thus permitting activation and/or synthesis of large number of enzymes (e.g., ACC oxidase)

and greater mobilization of storage reserves. Some of these events seemingly better prepare the seeds to resist or tolerate cold temperature.

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