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Temperature Effects on Imbibition and Germination of Cucumber (*Cucumis sativus*) Seeds

Paul Jennings[1] and Mikal E. Saltveit
Mann Laboratory, Department of Vegetable Crops, University of California, Davis CA 95616-8631

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***Abstract*. Unlike horticulturally mature fruit of 'Dasher II' and 'Poinsett 76' cucumbers (Cucumis sativus L.), two cultivars that differ significantly in their level of chilling tolerance, imbibing and germinating seeds of these two cultivars responded similarly to chilling temperatures (e.g., increases in fresh weight, time to radicle emergence, and root growth). 'Dasher II' and 'Poinsett 76' seeds were imbibed and germinated at 10 to 30C, and seeds germinated at 25C for 24 h were chilled at 2.5C for various durations. In comparison, seeds from an aged lot of 'Poinsett 76' seed (1989) responded very differently from the 1992 seed lots in all experiments. The chilling tolerance level of germinating 'Poinsett 76' seed varied with the seedling age as measured by resumption of root growth. Our results suggest that some factor that confers chilling tolerance is gradually lost during the early stages of germination following imbibition.**

Uniform germination and seedling establishment is crucial for the commercial growth of many agricultural crops. Species that originated in the tropics or subtropics are especially sensitive to chilling (i.e., physiological injury caused by exposure to nonfreezing temperatures below ≈10C) that often occurs under early season temperate conditions (Bennett et al., 1992; Bradow, 1990; Christiansen, 1968; Harrington and Kihara, 1960; Herner, 1990; Nelson and Sharples, 1980; Pollock and Toole, 1966; Sachs, 1977). Although low soil temperatures can be avoided in commercial practice by delaying planting, there is an economic incentive to achieve early season harvest or extend the growing season by planting under less than ideal conditions early in the season. The delay in planting to await more favorable temperature conditions can be compensated for by using transplants, but transplants of chilling sensitive crops are also susceptible to injury when exposed to chilling temperatures during production or after planting.

Sensitivity to chilling varies among the cultivars of many important agricultural crops, among the tissues in the plant, and among the age and developmental stage of the plant (Couey, 1982; Saltveit and Morris, 1990). All tissues in a cucumber plant and fruit are chilling sensitive (Cabrera et al., 1992; Saltveit, 1994). Exposure of horticulturally mature cucumber fruit to 2.5C for 8 days produced different levels of chilling injury in 'Dasher II' and 'Poinsett 76' fruit. 'Dasher II' fruit was significantly more tolerant to chilling than 'Poinsett 76' fruit based on the severity of pitting, the incidence of decay, the rate of ion leakage, the fresh and dry weight of the exudate, and the visual quality (Cabrera et al., 1992).

This study was undertaken to examine temperature effects on imbibition, germination, and chilling sensitivity of 'Dasher II' and 'Poinsett 76' seeds—two *Cucumis sativus* cultivars that previously differed significantly in the chilling tolerance of their fruit.

Materials and Methods

Plant material. Seeds of 'Dasher II' and 'Poinsett 76' were obtained from Peto Seed Co., Woodland, Calif. In addition, an aged sample of 'Poinsett 76' seeds, initially obtained from Peto Seed Co. in 1989 and held in a paper bag at room temperature and relative humidity, was used for comparison in some experiments.

Temperature effect on imbibition. One hundred dry seeds were weighed and placed in 9-cm plastic petri dishes over two layers of paper toweling wetted with 9 ml deionized water, which had been previously equilibrated at either 25 or 2.5C. At appropriate time intervals, the seeds were removed, blotted dry, and weighed.

Germination rate and cumulative germination. Seeds were placed in 5-cm tight-lid plastic petri dishes with two layers of paper toweling wetted with 3 ml deionized water. Each experiment was run in duplicate with 25 seeds per treatment. Temperature treatments were 10, 15, 20, 25, and 30C. At appropriate times, the seeds were examined and those which had a >3 mm radicle visible were counted as having germinated and removed. Cumulative germination (G), mean germination rates (i.e., the inverse of mean time to germinate), and time to 50% and 80% germination were determined from plots of the data.

Effect of chilling duration. Seeds were imbibed for 8 h at 25C in 9-cm plastic petri dishes with two layers of paper toweling wetted with 9 ml deionized water. The imbibed seeds were then transferred to paper toweling on a piece of water-saturated rock wool (12 × 29 × 2 cm), and covered with another layer of wet paper toweling. The rock wool was sandwiched between two plexiglass plates secured with rubber bands and oriented to a vertical position for an additional 16 h at 25C. Radicles were ≈0.2-cm long after this 24-h treatment. The germinated seeds were then placed in 9-cm plastic petri dishes with two layers of paper toweling wetted with 8 ml deionized water equilibrated at 2.5C and chilled at 2.5C for 0, 72, 96, 120, or 144 h. At the end of the chilling period, the germinated, chilled seeds were transferred to vertical rock wool slabs as previously described and incubated at 25C for up to 96 h.

Effect of stage of germination on chilling sensitivity. Seeds were imbibed in petri dishes for 8 h as previously described, before transfer to the rock wool sandwiches for the remainder of the imbibition period (e.g., 16, 20, 24, 28, or 32 h) at 25C. At the end of the respective imbibition period, the seeds were placed in plastic petri dishes with two layers of paper toweling wetted with 8 ml deionized water equilibrated at 2.5C and chilled at 2.5C for various durations (e.g., 0, 72, 96, 120, or 144 h). At the end of the chilling period, the germinated, chilled seeds were transferred to vertical rock wool slabs as previously described and incubated at 25C for up to 96 h.

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[1]Work done while on sabbatical at Univ. of California at Davis. Horticulture Dept., Kansas State Univ., Manhattan, KS 66506-4002

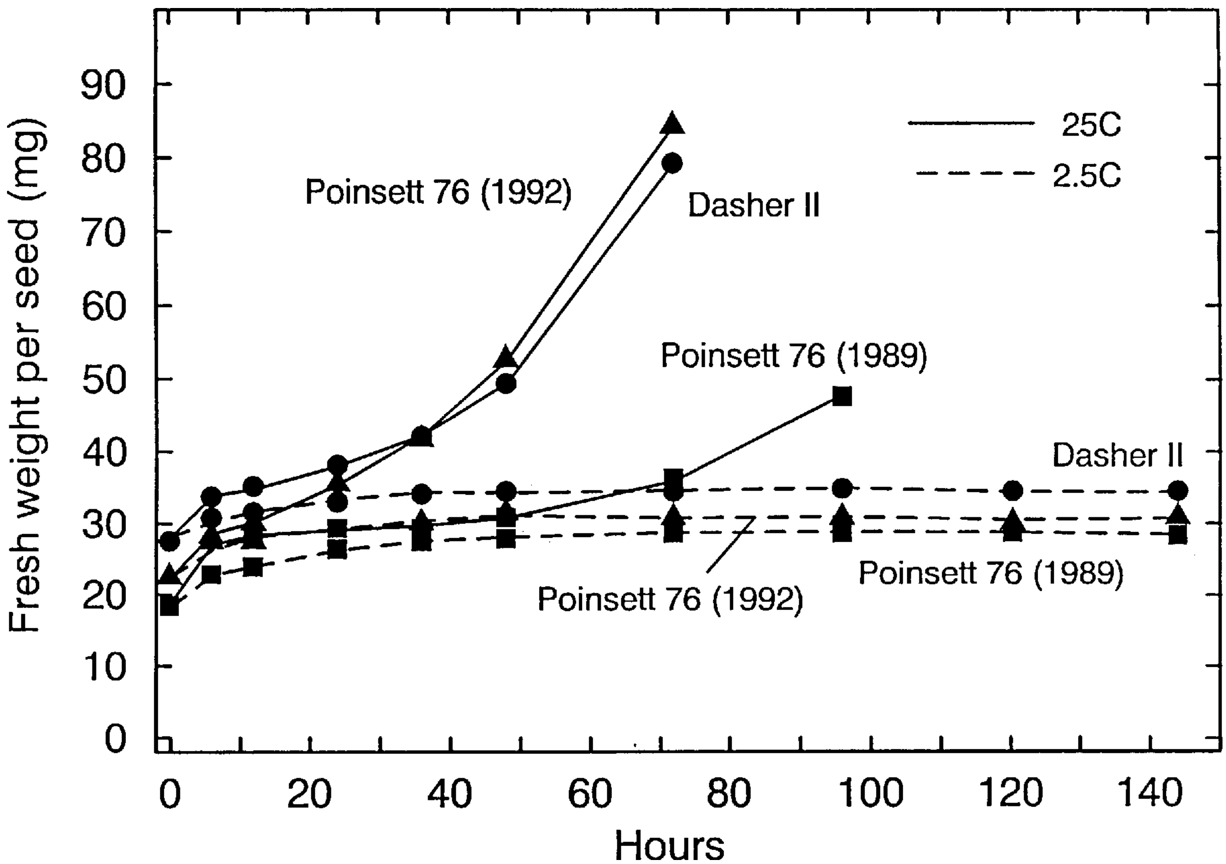

Fig. 1. Effect of imbibition temperatures of 2.5 and 25C on fresh weight changes during germination of 'Dasher II' and 'Poinsett 76' cucumber seed from 1992 and 'Poinsett 76' seed from 1989.

Table 1. Effect of germination temperatures on the time to 50% and 80% germination of 'Dasher II' and 'Poinsett 76' cucumber seeds from 1992 and 'Poinsett 76' seeds from 1989.

	Hours to 50% germination			Hours to 80% germination		
Temp	Dasher II	Poinsett 76		Dasher II	Poinsett 76	
(C)	(1992)	(1992)	(1989)	(1992)	(1992)	(1989)
15	44.0 a[x]	48.6 a	77.5 b	49.1 a	51.5 a	132.6 b
20	29.9 a	33.3 a	62.2 b	34.5 a	38.4 a	99.8 b
25	15.2 a	15.0 a	37.1 b	18.4 a	18.4 a	55.2 b
30	8.2 a	7.5 a	32.2 b	13.6 a	12.0 a	44.1 b

[y]Means in the same temperature (row) and germination percentage (columns) that are followed by the same letter are not significantly different at $P = 0.05$.

Results

Temperature effects on imbibition. Although the extent of imbibition at 2.5C, as measured by weight gain per seed, was slight and did not significantly increase after 20 h, the rate and extent of imbibition was similar for both 'Poinsett 76' and 'Dasher II' seed (Fig. 1). The rate and extent of imbibition at 25C was much greater than at 2.5C for all seed lots. The 1992 seed lots of 'Dasher II' and 'Poinsett 76' were similar to each other in their rate and extent of imbibition at 25C. The 1989 'Poinsett 76' seed lot became significantly different from the two 1992 seed lots after 24 h at 25C, but did not become significantly different from the seeds imbibed at 2.5C until 96 h. The time of radicle emergence at 25C was also very similar between the 1992 seed lots of the two cultivars; however, the 1989 'Poinsett 76' seed lot exhibited delayed radicle emergence.

Cumulative germination. When seeds were germinated over a range of temperatures from 15 to 30C, the time to 50% germination was similar for the 1992 seed lots of 'Poinsett 76' and 'Dasher II' (Table 1). The time to 50% germination for the 1989 'Poinsett 76' seed lot was statistically different from the two 1992 seed lots. The linear equation $84.55 - (2.64 \times \text{temperature})$ gave the time to 50% germination with an r^2 of 0.97 for the two 1992 seed lots. The equation for the 1989 seed lot was (time to 50% germination) = $124.70 - (3.22 \times \text{temperature})$ with an r^2 of 0.95. The two 1992 seed lots were also similar in the amount of time each took to reach 80% germination. The linear equation $88.24 - (2.61 \times \text{temperature})$ gave the time to 80% germination with an r^2 of 0.96 for the two 1992 seed lots. The equation for the 1989 seed lot was (time to 80% germination) = $222.47 - (6.20 \times \text{temperature})$ with an r^2 of 0.96.

Mean germination rate. The mean germination rate for the 1992 seed lots of 'Dasher II' and 'Poinsett 76' were similar at temperatures from 15 to 30C (Fig. 2). The mean germination rate at 5 and 10C was zero for all seeds. The 1989 seed lot of 'Poinsett 76' was significantly different from the two 1992 seed lots at 15 to 30C.

Effect of chilling duration. Exposing 'Poinsett 76' seeds (1992 seed lot) that had been germinated for 24 h at 25C to chilling at 2.5C for time periods up to 120 h, progressively reduced subsequent root growth at 25C (Fig. 3). When measured at 48 h after chilling, the reduction in root growth was linear ($r^2 = 0.98$) for up to 48 h of chilling. Measurements at 72 and 96 h were also linear ($r^2 = 0.97$ and 0.98, respectively) with duration of chilling for up to 72 and 96 h, respectively. In other words, the seeds appeared to recover from chilling and resume growth, and the length of time for recovery was proportional to the duration of chilling. In all cases,

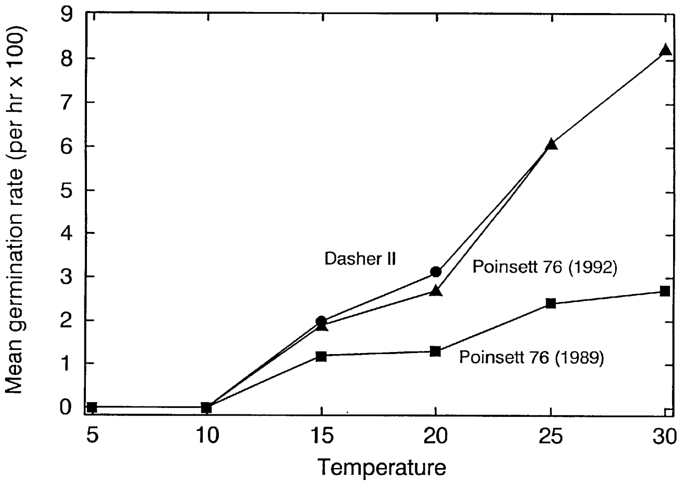

Fig. 2. Effect of temperature on mean germination rate of 'Dasher II' and 'Poinsett 76' cucumber seed from 1992 and 'Poinsett 76' seed from 1989.

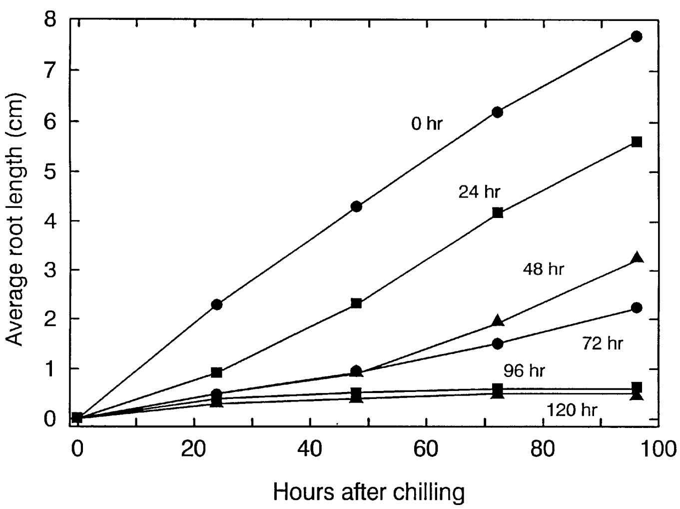

Fig. 3. Effect of duration of chilling at 2.5C on root growth of 'Poinsett 76' seed germinated at 25C for 24 h.

however, ≥96 of chilling caused irreversible damage and death of the root tip (Fig. 3).

Effect of stage of germination on chilling sensitivity. The degree of chilling sensitivity of 'Poinsett 76' seeds varied with seedling age (Fig. 4). Seeds were germinated at 25C for 16 to 32-h periods before being chilled at 2.5C for 72 to 144 h. As the age of the seedlings increased from 16 to 28 h, they became more chilling sensitive and the root growth from 24 to 96 h after chilling steadily declined. The moderate chilling stress of exposure to 2.5C for 72 or 96 h had the greatest effect on reducing subsequent root growth in 28-h-old seedlings. Seedlings 4 h older or younger than 28 h were significantly less chilling sensitive. More severe levels of chilling stress, imposed by exposure to 2.5C for 120 and 144 h, however, caused a progressive decline in root growth with seedling age.

Discussion

Imbibition of cucumber seeds appears to be completed between 12 and 16 h at 25C and by 24 to 36 h at 2.5C. Unlike cottonseed, which are reported to be killed when hydrated at 5C (Christiansen, 1968), cucumber seeds may be imbibed for at least 6 days at 2.5C and still retain their ability to germinate normally

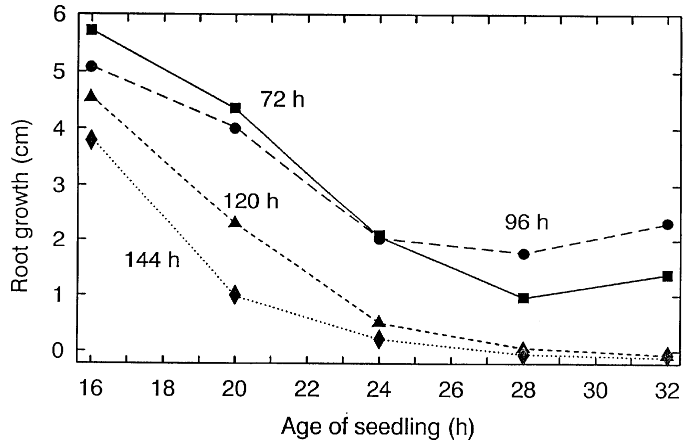

Fig. 4. Effect of duration of germination of 'Poinsett 76' cucumber seeds at 25C on the increase in root length from 24 to 96 h after the various aged germinated seeds were chilled at 2.5C for 72, 96, 120, or 144 h.

(data not shown).

Even though a difference in chilling sensitivity of the fruit existed between 'Poinsett 76' and 'Dasher II' (Cabrera et al., 1992), no differences in chilling sensitivity were detected during seed germination under chilling conditions. The mean germination rates for the two cultivars were indistinguishable over the temperature range of 10 to 30C.

Results with older 'Poinsett 76' seedlings have also shown chilling sensitivity when exposed for 5 to 6 days at 10 and 15C as determined by subsequent root growth (Bradow, 1990).

Herner (1990) reported that many plant species not susceptible to chilling injury during imbibition become chilling sensitive at the time of radicle emergence. Our results confirm this observation for cucumber seeds imbibed at 25C for 24 h. Radicles are irreversibly damaged when exposed to 2.5C for 96 to 120 h. Harrington and Kihara (1960) reported similar responses with muskmelon germinated under chilling conditions and described a sunken spot near the tip of the radicle and collapse of tissue in this area. However, there also appears to be a gradual increase in susceptibility to chilling with increasing time of germination between 16 and 32 h. In a study with germinating pepper seeds, Irwin and Price (1983) showed that seedlings with longer radicles had reduced emergence when chilled at 0C for 4 days.

We have shown that a longer chilling treatment is required to reduce growth and cause irreversible damage in 16-h-old germinated cucumber seeds than in seeds germinated for 28 or 32 h. These results suggest that some factor that confers chilling tolerance is gradually lost during the early stages of germination following imbibition.

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