Screening USDA Lettuce (*Lactuca sativa* L.) Germplasm for Ability to Germinate under Cold Conditions

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Abstract. Low temperatures can slow down emergence, decrease weed competitiveness, and lead to uneven crop maturity in direct-seeded crops such as baby leaf lettuce (*Lactuca sativa* L.). In this study, seeds of 103 single-parent lineage, homozygous lettuce accessions (53 cos and 50 leaf type) from the USDA National Plant Germplasm System (NPGS) and six commercial standard lettuce cultivars (three cos and three leaf type) were evaluated in replications for percent germination after 7 and 10 days at 5 °C in a germination chamber. Cos and leaf types were selected for this study as they are most commonly used for baby leaf lettuce production. Differences were observed among entries in percent seeds germinated after both 7 and 10 days. Overall, an average of 68% of seeds germinated after 7 days and 94% germinated after 10 days. Although several NPGS accessions had higher percent germination than the commercial cultivars, the average percent germination was not statistically different between the two seed sources at 7 or 10 days. Percent germination also did not differ between entries of cos and leaf type after 7 or 10 days. Similarly, no difference in percent germination between entries of dark and white seed color was observed after 7 or 10 days. No relationship between 100 seed weight and percent germination was observed after 7 days ($r^2 = 0.07$) or 10 days ($r^2 = 0.13$). Thus, lettuce seed type, color, and 100 seed weight do not appear to be good predictors of germination under cold conditions in lettuce. The accessions with the highest percent germination after 7 days at 5 °C have the potential to be used for the development of new lettuce cultivars suitable for an extended, early season production in temperate climates when soil temperature is lower than optimal for lettuce germination. Further studies are needed to assess the ability of these accessions to germinate rapidly under cold field soil conditions.

Lettuce is a cool-season crop that germinates optimally at an average temperature of 18 °C (Lafta and Mou, 2013). Established

lettuce plants can survive a wide range of environmental conditions including moderate frost and daily temperature down to –12 °C and as high as 35 °C (Rubatzky and Yamaguchi, 1997; Shibutani and Kinoshita, 1970). Although there does not appear to be any permanent impact on crop health and quality under these adverse conditions, lettuce plants display a reduced growth rate relative to crop performance under optimal temperatures (Cantliffe, 1989; Rubatzky and Yamaguchi, 1997). In contrast, newly planted lettuce seeds and developing seedlings are less tolerant to extreme temperatures than mature lettuce plants. At 20 °C, lettuce seed germination occurs on average 2 d

after planting at 1 cm depth, whereas at 5 °C emergence occurs an average of 15 d after planting, and at 3 °C or lower, lettuce seed fails to germinate (Wagenvoort and Bierhuizen, 1977). Similarly, lettuce seed germination and emergence is suppressed at temperatures over 30 °C for most cultivars (Mayberry, 2003), though genotypes, specifically *L. serriola*, have been identified, which can germinate at 35 °C (Argyris et al., 2008).

Quick emergence of seedlings has long been the goal of horticulturists as a means of optimizing stand uniformity and maturity in direct-seeded crops (Fromme et al., 2014; Seale and Cantliffe, 1987). Low soil temperatures can increase the time to emergence, which leads to uneven crop maturity (Herner, 1986). This is problematic in a production system that uses once-over harvesting, as with mechanically harvested baby leaf lettuce. Slow stand establishment also results in a lengthened amount of time to complete canopy formation, which leads to increased weed emergence and growth (Légère and Schreiber, 1989).

Slow and inconsistent germination under cold soil temperature conditions is a primary barrier to extending the production season for lettuce in temperate regions. California and Arizona are the main production areas of leafy green salad crops, producing 96% of all lettuce grown in the United States, with 70% of baby leaf salad crops produced in the Salinas Valley (City of Salinas EDO, 2013; USDA, 2014). The mild winter climate in these regions makes it possible to produce quality lettuce year-round (Smith et al., 2009). In contrast, lettuce production in temperate regions such as northwest Washington is limited to May through September, when soil temperature and moisture conditions are most favorable for crop emergence and growth. Spring soil temperature in northwest Washington averages 10 °C (AgWeatherNet, 2014), and lettuce seeds emerge slowly and unevenly in these conditions.

Seed vigor comprises those properties that determine the potential for rapid, uniform emergence and development of normal seedlings under a wide range of field conditions (McDonald, 1993). Seed vigor is based on seed quality, environmental and maternal effects during seed crop production, as well as the genetic constitution of seeds which establishes their maximum physiological potential (Sun et al., 2007). Seed vigor encompasses several interrelated characteristics including rate and uniformity of germination and seedling growth, ability of seeds to germinate and emerge under unfavorable environmental conditions, and retention of the ability to germinate after storage. Seed vigor has already been shown to have a genetic component in other direct-seeded crops such as wheat (*Triticum aestivum* L.) (Rebetzke et al., 2004), cole crops including a Chinese kale (*Brassica oleracea* L.) × calabrese broccoli (*B. oleracea* L.) cross, a Brussels sprout (*B. oleracea* L.) × cauliflower cross (*B. oleracea* L.) (Finch-Savage et al., 2010), rice (*Oryza sativa* L.) (Redona and

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Mackill, 1996), and sugar beet (*Beta vulgaris* L.) (De Los Reyes et al., 2003).

Seed morphological traits such as size and seedcoat color have been linked to seed vigor and emergence in many crop species, including lettuce. For example, watermelon (*Citrullus lanatus* var. *lanatus* L.) seed vigor, time to emergence, and viability can be determined according to seedcoat color, with dark seeds having better performance than light seeds (Mavi, 2010). In dry pea (*Pisum sativum* L.), single genotypes produce light, medium, and dark seed, and the color of the seed is linked to characteristics such as germination behavior, seedling salinity tolerance, and resistance to soilborne fungi, with dark seeds having the highest vigor under high salinity conditions and in the presence of soilborne pathogens (Atak et al., 2008). Lettuce seedcoat color heredity is such that varieties and genotypes can be classified as either dark or white seeded, with little to no variation in seed color observable within a genotype (Penaloza et al., 2005). Dark-seeded lettuce has been observed to possess higher percent germination and seed vigor as well as a lower pathogen susceptibility than white-seeded lettuce under 20 °C conditions (Penaloza et al., 2005). Seed size and weight are associated with seed germination for maize (*Zea mays* L.), where, within a genotype, larger, heavier seeds generally have higher germination percentage and vigor than small seeds (Batistella et al., 2002). The relationship between seed weight and percent germination in lettuce has not been confirmed.

This experiment compared final germination at 5 °C for lettuce germplasm from the NPGS with standard cultivars used for baby leaf lettuce production by individual genotype, lettuce type (cos and leaf), seedcoat color, and seed weight. Identifying lettuce accessions from the NPGS collection that germinate rapidly under cold temperature conditions could lead to development of new cultivars with improved seedling vigor, which in turn could improve extended season production of direct-seeded lettuce in temperate regions.

Materials and Methods

Seed. Single-parent lineage, homozygous lettuce accessions were obtained from the USDA NPGS Western Region Plant Introduction Station (WRPIS), Pullman, WA. From the pure-line lettuce special collection of 298 lines, each of which is homozygous for all 322 single-nucleotide polymorphism markers genotyped by a high-throughput assay (Kwon et al., 2013), a total of 103 cos and leaf lettuce types were chosen for evaluation in this study as these are the lettuce types most commonly grown for baby leaf lettuce production. All seed obtained from the USDA NPGS was grown from a planting in 2011 (71 accessions) or 2012 (32 accessions) at the USDA Agricultural Research Service Central Ferry Farm, Washington. Seed was harvested in September or October in

both years and was stored under controlled temperature and relative humidity (RH) conditions (4 °C, 20% RH) at the WRPIS seed storage facility in Pullman. The accessions included 53 cos accessions (14 dark and 39 white seeded) and 50 leaf type accessions (27 dark and 23 white seeded), for a total of 41 dark- and 62 white-seeded accessions. Most accessions had greater than 90% germination in a germination test performed at 20 °C. The six commercial standard cultivars (packaged for 2014; Johnny's Select Seeds, Winslow, ME) included in this study were three cos cultivars Parris Island, Coastal Star, and Defender (all white) and three-leaf cultivars Black Seeded Simpson, Two Star, and Waldmann's Green (all dark). These cultivars were chosen as they are commonly grown for commercial baby leaf lettuce production. Commercial standard cultivars were tested for germination at 20 °C from Dec. 2013 through Apr. 2014 and all had a final germination exceeding 94%.

Assay methodology. At Washington State University Northwestern Washington Research and Extension Center in Mount Vernon, WA, lettuce seed germination was evaluated in a germination chamber (Hoffman Model SG 30; Hoffman Manufacturing Inc., Jefferson, OR) with a 12 h light:12 h dark cycle under fluorescent light (180 $\mu\text{mol}\cdot\text{s}^{-1}\cdot\text{m}^{-2}$). For this study, the temperature was 5 ± 1 °C, and was recorded every 15 min throughout the study (HOBO data logger; Onset Computer Corporation, Bourne, MA). This study included three replicates of 50 seeds because of limited quantity of available seed. For each replicate, 50 seeds of each accession were placed into an acrylic container (5.25 × 5 inches; Hoffman Manufacturing, Jefferson, OR) lined with Steel Blue blotter paper (Anchor Paper, Saint Paul, MN) and saturated with deionized water. Germination boxes were covered and placed randomly in the germination chamber. Number of seeds with normal germination was recorded for each accession at 7 and 10 d after placement in the germination chamber. Normal germination was determined according to the Association of Official Seed Analysts (AOSA) definition, which includes a normally developed hypocotyl, root tip, and primary root; and two normally developed cotyledons free of necrosis, disease, or discoloration (AOSA, 2009).

This experiment was carried out in two stages with the same germination equipment and methods used for both. First, the complete set of 103 NPGS germplasm accessions were assayed from 21 to 30 Sep. 2013. Second, a subset of 18 lettuce accessions (nine cos and nine leaf) plus six commercial standards (three cos and three leaf) were assayed from 7 to 16 Feb. 2015. The accessions included in the second assay were selected based on their percent germination at 7 d in the first assay; three cos and three leaf accessions were selected from each of the germination groups: high (80% to 100%), moderate (45% to 65%), or low (0% to 25%). For each accession, five sets of 100 seeds

were weighed to obtain the average 100 seed weight. For commercial standards, 100 seed weight was calculated from the average number of seeds per pound (data obtained from Johnny's Selected Seeds) for the specific seed lot of each commercial standard cultivar evaluated in this study. The 100 seed weight was compared with germination percentage at 7 and 10 d, and seed weight was also compared between lettuce types and seed colors.

Statistical analysis. To evaluate consistency between the first and second assays, the percent germination at 7 and 10 d of the 18 accessions included in both assays were subjected to a paired *t* test using PROC TTEST in SAS (Statistical Analysis System Version 9.3 for Windows; SAS Institute, Cary, NC). When no differences were detected between the performance of these 18 accessions at 7 or 10 d ($P = 0.45$ and 0.13 , respectively), data from the two assays were combined and "assay" (first and second) was incorporated into the statistical model as separate blocks. Combined data were subjected to analysis of variance (ANOVA) using PROC MIXED in SAS (Version 9.3 for Windows). Data were analyzed by entry, lettuce type (cos or leaf), and seedcoat color (dark or white). Raw data did not meet analysis assumptions of normality or homogeneity of variance; therefore, an attempt was made to identify an appropriate transformation meeting these assumptions using the range method described by Kirk (1982). When no transformation satisfied these assumptions, a nonparametric ANOVA was performed as described above after converting data into ranks using PROC RANK in SAS (Version 9.3 for Windows). Fisher's least significant difference test was used to compare treatment means for significant differences. No interactions were detected between main effects. Correlation between 100 seed weight and final germination at 7 and 10 d was determined with a simple linear regression model using PROC REG in SAS (Version 9.3 for Windows).

Results

Percent germination differed among entries at both 7 and 10 d (both $P = 0.0001$). Of the 63 entries with high final germination (entries with the highest means separation level) at 7 d, 60 were accessions and 3 were commercial standards (Table 1). Of the accessions with the highest final germination, 13 (seven cos and six leaf) had 100% germination, while commercial leaf-type cultivars Black Seeded Simpson (91.7%) and Defender (91.1%), and cos-type cultivar Two Star (83.7%) were statistically equivalent. Of the 26 entries with medium germination (entries with neither the highest nor the lowest means separation level), 24 were accessions and 2 were commercial standards ('Waldmann's Green', 60.4% and 'Parris Island', 48.4%). Of the 20 entries with low germination (entries with the lowest means separation level), 19 were accessions and one

Table 1. Germination (%) of 109 lettuce germplasm entries after 7 and 10 d at 5 °C.^z

Entry ^y	Lettuce type	Seed coat color	Mean germination (%) ^x			
			7 d		10 d	
PI 169494-0	Leaf	Dark	100	a ^w	100	ab
PI 175737-0	Leaf	Dark	100	a	100	ab
PI 566589-0	Leaf	Dark	100	ab	100	ab
PI 278108-1	Cos	Dark	100	ab	100	ab
PI 342552-1	Cos	Dark	100	ab	100	ab
PI 175736-0	Leaf	White	100	a-c	100	ab
PI 342474-1	Leaf	White	100	a-c	100	ab
PI 344366-1	Leaf	White	100	a-c	100	ab
PI 491025-0	Cos	White	100	a-d	100	ab
PI 491056-0	Cos	White	100	a-d	100	ab
PI 181946-1	Cos	White	100	a-d	100	ab
PI 278081-1	Cos	White	100	a-d	100	a
PI 288244-3	Cos	White	100	a-d	100	a
PI 182210-1	Leaf	Dark	98.9	a-d	98.9	a-c
PI 164937-0	Leaf	White	98.7	a-d	100	ab
PI 536764-0	Leaf	Dark	98.3	a-d	98.3	a-e
PI 342541-4	Cos	Dark	97.6	a-d	97.9	a-e
PI 142871-0	Cos	Dark	97.5	a-e	100	a ^w
PI 220665-1	Cos	Dark	97.3	a-f	99.3	ab
PI 278099-1	Cos	White	96.7	a-f	100	ab
PI 491042-0	Cos	White	95.6	a-g	95.6	a-f
PI 491053-0	Cos	White	94.9	a-g	95.9	a-f
PI 178923-1	Leaf	Dark	94.3	a-g	94.3	a-f
PI 536821-0	Cos	Dark	93.6	a-h	97.2	a-e
PI 169493-0	Cos	White	93.6	a-h	97.9	a-e
PI 491040-0	Cos	White	92.3	a-h	95.2	a-f
PI 491212-0	Cos	White	92.3	a-h	96.5	a-f
Black Seeded Simpson	Leaf	Dark	91.7	a-h	100	ab
PI 491080-0	Leaf	White	91.6	a-h	97.9	a-e
Defender	Cos	White	91.1	a-i	100	ab
PI 171666-0	Cos	Dark	90.9	a-i	97.9	a-e
PI 491032-0	Cos	White	90.8	a-i	97.7	a-e
PI 536700-0	Leaf	Dark	88.3	a-i	96.5	a-f
PI 617943-1	Leaf	White	88.3	a-i	95.9	a-f
PI 491031-0	Cos	Dark	87.8	a-i	98.3	a-e
PI 278080-1	Leaf	White	87.6	a-i	95.9	a-f
PI 536794-0	Cos	White	86.9	a-i	97.2	a-e
PI 381933-0	Cos	Dark	84.3	a-i	97.2	a-e
PI 177419-0	Leaf	White	84.3	a-i	95.9	a-f
PI 271476-1	Leaf	Dark	84	a-k	97.3	a-e
Two Star	Leaf	Dark	83.7	a-k	100	ab
PI 536699-0	Leaf	Dark	83.3	a-l	99.3	ab
PI 368630-1	Cos	White	82.9	a-m	96.5	a-f
PI 289016-2	Leaf	White	82.3	a-m	97.9	a-e
PI 358033-1	Cos	White	82.3	a-m	93.9	a-f
PI 536792-0	Leaf	Dark	81.6	a-m	97.9	a-e
PI 617944-0	Leaf	White	80.9	a-n	93.9	a-f
PI 176980-0	Cos	White	77.6	a-o	92.5	a-g
PI 172918-0	Cos	White	75.6	a-o	97.9	a-e
PI 491078-0	Leaf	Dark	74.9	a-o	95.2	a-f
PI 617958-1	Leaf	White	74.9	a-o	91.9	a-g
PI 176588-0	Cos	Dark	74.3	a-o	89.2	c-h
PI 578884-0	Leaf	White	73.6	a-o	97.2	a-e
PI 176579-2	Cos	White	73.6	a-o	97.9	a-e
PI 491039-0	Cos	White	71.6	a-o	97.2	a-e
W6 298171	Leaf	Dark	70.9	a-o	98	a-e
PI 491084-0	Cos	White	70.9	a-p	96.5	a-f
PI 358001-1	Leaf	White	70.3	a-p	82.5	g-j
PI 278087-0	Cos	White	70.3	a-p	97.2	a-e
PI 491045-0	Cos	White	70.3	a-p	89.2	c-h
PI 491213-0	Cos	White	70.3	a-q	97.2	a-e
PI 508477-1	Leaf	Dark	69.6	a-r	97.9	a-e
PI 146078-7	Leaf	White	68.9	a-r	97.9	a-e
PI 617958-3	Leaf	Dark	67.6	b-s	91.9	a-g
PI 491034-0	Cos	White	66.9	b-s	97.2	a-e
PI 229762-0	Cos	Dark	65.6	c-s	79.2	h-j
PI 491008-0	Cos	White	65.6	c-s	74.5	jk
PI 169509-1	Cos	White	65.6	c-s	92.5	a-g
W6 3687	Leaf	Dark	64.9	d-s	64.9	kl
PI 146078-1	Leaf	White	64.9	d-s	97.2	a-e
PI 491049-0	Cos	White	62.9	d-s	96.5	a-f
PI 491215-0	Cos	White	62.3	e-t	82.5	g-j
PI 358021-0	Leaf	Dark	61.6	f-t	91.9	a-g

(Continued on next page)

was a commercial standard ('Coastal Star', 31.1%). At 10 d, 20 entries (16 accessions and four commercial standards) achieved 100% germination (Table 1). Of these entries, 11 were cos and nine were leaf type. Of the 21 entries with the lowest percent germination (all were accessions), 12 were cos and nine were leaf type.

Average percent germination for all accessions was 68% after 7 d and 94% after 10 d. Average percent germination of accessions did not differ from the commercial standards after 7 d (68% both; $P = 0.77$) and 10 d (accessions = 94%, commercial standards = 99%; $P = 0.29$). No difference in percent germination was observed between cos and leaf lettuce types at 7 d (cos = 70%, leaf = 65%; $P = 0.38$) or 10 d (cos = 95%, leaf = 94%; $P = 0.66$). In addition, no difference was observed between entries with dark and white seedcoats at 7 d (dark = 70%, white = 66%; $P = 0.13$) or 10 d (dark = 95%, white = 94%; $P = 0.07$). No relationship was observed between 100 seed weight and lettuce type ($P = 0.56$), 100 seed weight and seed color ($P = 0.92$), or 100 seed weight and seed source ($P = 0.70$) (Table 2). Although statistically significant, there was no meaningful relationship observed between percent germination and 100 seed weight at either 7 d ($r^2 = 0.07$, $P = 0.007$) or 10 d ($r^2 = 0.13$, $P = 0.005$).

Discussion

The seed assessed in this experiment was produced over two growing seasons, and commercial cultivar seed was produced at a different location than the NPGS accession seed. Therefore, the observed seed performance is potentially affected by genotype by environment interactions. After 7 d at 5 °C, 13 NPGS accessions (seven cos and six leaf) had 100% germination, whereas the commercial cos and leaf cultivars with the highest final germination had 91.1% and 91.7% germination, respectively. Higher germination percentage at 7 d may result in faster stand establishment and canopy formation under field conditions (Orzolek, 1991). The 13 NPGS accessions with the highest percent germination at 7 d may be useful for developing new cultivars with high seed vigor in cold soil conditions for use in extended season production of direct-seeded lettuce.

In this study, lettuce type, seed weight, and seed color were not reliable predictors for final germination under cold temperature conditions. Large seed size and high seed weight have been linked to high seed vigor in crops such as wheat and corn (Hoy and Gamble, 1985; Lafond and Baker, 1986). This relationship has not been consistently observed in lettuce and no relationship was observed in this experiment.

Temperature conditions in this experiment did not favor the growth of any disease inoculum that may have been present on seeds, so no conclusions can be drawn from this study on the linkage between seedcoat color, size, or lettuce type on pathogen

Table 1. (Continued) Germination (%) of 109 lettuce germplasm entries after 7 and 10 d at 5 °C.^z

Entry ^y	Lettuce type	Seed coat color	Mean germination (%) ^x			
			7 d		10 d	
Waldmann's Green	Leaf	Dark	60.4	f-t	97.5	a-e
PI 536826-0	Leaf	Dark	59.6	g-t	97.2	a-e
PI 358039-1	Cos	White	57.6	h-u	97.2	a-e
PI 615060-1	Leaf	Dark	54.9	i-u	95.2	a-f
PI 278110-2	Leaf	White	54.9	i-u	97.9	a-e
PI 176578-0	Leaf	Dark	54.3	j-t	99.7	ab
PI 491062-0	Leaf	White	54.3	j-t	87.7	f-i
PI 278094-0	Cos	Dark	48.9	j-w	95.2	a-f
Parris Island	Cos	White	48.4	j-w	99.5	ab
PI 601060-0	Leaf	Dark	48.3	j-w	96.5	a-f
PI 583877-0	Leaf	White	46.9	k-x	97.9	a-e
PI 172916-1	Leaf	Dark	46.3	l-x	95.9	a-f
PI 536845-0	Leaf	White	45.6	l-x	63.9	kl
PI 285657-0	Cos	White	45.3	m-x	95.3	a-f
PI 491070-0	Leaf	White	43.6	n-v	97.9	a-e
PI 278109-1	Leaf	White	43.3	n-y	80	h-j
PI 491079-0	Leaf	Dark	33.6	o-w	89.2	c-h
PI 578883-0	Leaf	White	32.9	p-y	96.5	a-f
PI 278080-2	Leaf	White	31.6	q-y	96.5	a-f
Coastal Star	Cos	White	31.1	r-y	100	ab
PI 491065-0	Leaf	Dark	30.9	s-y	89.2	c-h
PI 491022-0	Cos	Dark	30.9	s-y	97.9	a-e
PI 342479-1	Cos	White	25.6	s-y	78.5	h-j
W6 29841	Cos	White	21.8	t-y	87.9	e-h
PI 169509-4	Cos	White	20.9	u-y	85.9	f-i
PI 140394-0	Cos	White	20.7	v-y	91.7	c-g
PI 175735-0	Leaf	White	17.6	v-y	96.5	a-f
PI 171665-1	Cos	White	17.3	v-y	80	h-j
PI 344367-1	Leaf	Dark	16.3	v-y	95.9	a-f
PI 278100-0	Cos	White	16.3	v-y	88.5	d-h
PI 274373-1	Leaf	White	15	w-y	58.7	lm
PI 342524-1	Leaf	Dark	7.6	xy	95.9	a-f
PI 491072-0	Leaf	Dark	6.9	y	63.9	kl
PI 342523-1	Leaf	Dark	6.9	y	93.2	a-g
PI 278072-0	Cos	White	6.9	y	85.9	f-i
PI 577120-0	Cos	White	6.9	y	93.9	a-f

^zLettuce seed (three replicates, n = 50 per entry) screened at 5 ± 1 °C temperature, 100% humidity, and 12 h light: 12 h dark in a Hoffman Model SG 30 germination chamber in acrylic containers (5.25 × 5 inches; Hoffman Manufacturing, Jefferson, OR) on steel blue blotter paper (Anchor Paper, St. Paul, MN).

^yPI and W6 lines obtained from the U.S. Department of Agriculture (USDA) National Plant Germplasm System (NPGS); commercial standard cultivars (bold) obtained from Johnny's Selected Seeds, Winslow, ME.

^xMean percent germination as defined by Association of Official Seed Analysts (AOSA) definition of normal lettuce seed germination of three replicates of n = 50 seeds per entry measured after 7 or 10 d in 5 °C conditions.

^wMean separation of ranked mean percent germination ($P = 0.0001$) as determined by a Fisher's least significant difference test. Ranked means with at least one letter in common are not significantly different.

Table 2. Single-effect means for seed color and lettuce type, and interaction with 100 seed weight.

Effect	100 seed wt (g) ^z
Seed color	
Dark	0.1029 ^y
White	0.1025
<i>P</i> value	0.92
Type	
Cos	0.1034
Leaf	0.1020
<i>P</i> value	0.56
Seed source	
Accession	0.1026
Standard	0.1028
<i>P</i> value	0.70

^zNo transformation satisfied the assumptions of normality for analysis of variance; therefore data were nonparametrically transformed using PROC RANK (SAS version 9.3 for Windows; SAS Institute, Cary, NC).

^yAll means presented are nontransformed.

under optimal field conditions for lettuce (Contreras and Barros, 2005; Smith et al., 1973). Field trials are needed to determine if accessions with the highest percent germination under cold germination chamber conditions (5 °C) will also have higher percent germination under cold field soil conditions.

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susceptibility. Future research might test for a relationship between seedcoat color and soilborne pathogen susceptibility in lettuce, similar to the relationship between these factors observed in dry green pea (Atak et al., 2008). Resistance to damping-off caused by *Pythium ultimum* and other fungal pathogens would be of particular interest to baby leaf lettuce growers who are direct-seeding into cool, moist soils. Damping-off is an economically significant problem in baby leaf lettuce production as high seedling mortality interferes with canopy formation and reduces yield (Kuepper et al., 2002). This disease affects baby leaf lettuce in cold soils especially, as the extended period in which the plant remains in the seedling stage under cold conditions results in a longer window of pathogen susceptibility.

Percent germination measured by the AOSA germination test (20 °C) has been shown to correlate with emergence rate

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