

## Intraspecific Variation in Seed Characteristics of Powell Amaranth (*Amaranthus powellii*) from Habitats with Contrasting Crop Rotation Histories

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The objectives of this research were to characterize the extent of intraspecific variation in seed characteristics of Powell amaranth and to evaluate whether such variation was correlated with crop rotation history of the collection sites. We compared characteristics of seeds originating from dairy farms with a corn–alfalfa crop rotation history with seeds originating from farms with a history of intensive vegetable production. We hypothesized that (1) multiple years of perennial alfalfa would select for greater seed dormancy and longevity in seeds of the summer annual Powell amaranth, (2) earlier spring planting dates of corn and alfalfa compared with most vegetable crops would select for earlier emergence, and (3) greater competition and lower soil moisture in the nonirrigated corn–alfalfa rotation would select for greater seed size. Seeds from 10 to 20 plants from each of 10 farms from each habitat were collected in the fall of 2002 and 2003 in central New York. To control for maternal effects on seed dormancy, a second generation of seeds was produced from plants grown under common greenhouse conditions. Germination in petri dishes was greater for second-generation seeds from vegetable farms (46%) than for those from dairy farms (32%). Total emergence following overwinter burial in the field was greater for seeds originating from dairy farms (62%) compared with those from vegetable farms (52%). Neither seed weight nor the rate of emergence varied by habitat of origin. Our results suggest that perennial alfalfa in dairy rotations may have selected for greater dormancy and longevity of Powell amaranth seeds. The large intraspecific variation in seed characteristics observed, underscores the importance of considering multiple populations when making comparisons of germination characteristics across biotypes (e.g., resistant vs. susceptible) or species, or when developing and interpreting models of weed emergence or weed population dynamics.

**Nomenclature:** Powell amaranth = green pigweed, *Amaranthus powellii* S. Wats. AMAPO; alfalfa, *Medicago sativa* L.; corn, *Zea mays* L.

**Key words:** Seed dormancy, microevolution, emergence, germination, intraspecific variation, ecotype.

Seed characteristics, including persistence, dormancy, and the timing of emergence, play a critical role in determining the population dynamics of weeds, and hence the long-term implications of different management practices (Cousens and Mortimer 1995). For example, the decision of how much effort and expense to devote to preventing weed seed production, depends critically on the likely fate of those seeds in the soil seed bank (Bussan and Boerboom 2001; Cardina et al. 1995). Likewise, the optimal timing of cultivation or likely success of stale seed bed practices depends on the timing of weed emergence (Forcella 1998).

Improved understanding of intraspecific variation in seed characteristics is important for defining the scope of inference from results of weed management studies, and may be useful for efficiently adapting weed management strategies to different cropping systems. Species-level assumptions about seed characteristics are central to models underlying decision support tools like WeedCast (Forcella 1998) or WEEDSIM (Swinton and King 1994). Because intraspecific variation in seed characteristics can be large, the robustness of such models across sites may be limited (Grundy 2003). Identification of habitat-correlated differences in seed characteristics could be useful for improving such models and facilitating more efficient weed management choices.

Intraspecific variation is also key to understanding microevolution, i.e., genetic differences among populations reflecting the past actions of selection and gene flow. Variation among populations is also a prerequisite for further adaptive change in response to new selective challenges such as global warming (Rice and Emery 2003). Therefore, characterization of intraspecific variation may provide insights into

the rate at which weeds can adapt to management practices, and hence the impact of those management practices on the abundance of a species (Clements et al. 2004; Mayor and Dessaint 1998; Menalled et al. 2001; Sosnoskie et al. 2006).

Although large intraspecific variations in seed characteristics have been shown for numerous species, studies exploring possible evolutionary explanations for such differences are less common, particularly in agroecosystems (Baskin and Baskin 1998). Several studies support the hypothesis that more dormant, longer-lived seeds are adaptive in situations where large temporal gaps occur between opportunities for seed production (Barrett and Wilson 1983; Brainard et al. 2006; Naylor and Jana 1976). Other studies suggest that large seeds may be adaptive under conditions of greater competition, since they increase the ability of seedlings to obtain light, nutrients, or water (Linhart 1974; Salisbury 1942; Tilman 1988). After controlling for maternal environmental effects, McWilliams et al. (1968) observed a lower base temperature for germination and greater seed weight for seeds of populations of redroot pigweed (*A. retroflexus* L.) originating from more northern latitudes. Using the same data set, Schimpf (1977) found a negative correlation between seed weight and moisture availability of the habitat of origin and hypothesized that larger seed size was advantageous in habitats with low moisture, because it hastened seedling root penetration to depths where moisture was more reliable.

Powell amaranth and its close relatives are pervasive and troublesome weeds in a wide range of crops throughout the world (Costea et al. 2004). Powell amaranth can reduce yields in a variety of crops including sugarbeet (*Beta vulgaris* L.) (Schweizer and Zimdahl 1984), broccoli (*Brassica oleracea* L. var. *italica*) and kale (*Brassica oleracea* L. var. *acephala*) (Brainard and Bellinder 2001), and soybean [*Glycine max* (L.) Merr.] (Dieleman et al. 1995). Management of Powell amaranth is complicated by the existence of biotypes resistant

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to triazine, phenylurea, and acetolactate synthase (ALS)-inhibitor herbicides (Costea et al. 2004; Tardif et al. 2006). In New York state, Powell amaranth is the dominant *Amaranthus* species (Senesac 1985) and is prevalent both on vegetable farms characterized by intensive production of annual crops and on dairy farms that rotate annual (e.g., corn) and perennial (e.g., alfalfa) crops. *Amaranthus* species can produce large quantities of long-lived seeds and hence often constitute the most numerous species in the seed banks of agricultural soils (e.g., Cardina et al. 1991; Mohler and Callaway 1995; Schweizer and Zimdahl 1984).

Frost and Cavers (1975) found large variation in the seed dormancy of six different seed lots of Powell amaranth but did not examine potential sources of that variation. In addition to potential genetic differences, maternal environmental conditions including competitive environment (Brainard et al. 2005a) and nutrient status (Brainard et al. 2005b) contribute to variability in Powell amaranth seed characteristics. After controlling for maternal environmental effects, Brainard et al. (2006) found greater seed germination and emergence for Powell amaranth seeds originating from organic compared to conventional vegetable farms and speculated that more effective control of Powell amaranth in certain rotational crops (e.g., sweet corn) in herbicide-based systems may have selected for greater seed dormancy.

The present study builds on this work by examining the effects of different crop rotation sequences on Powell amaranth seed characteristics. The objectives of this research were to characterize the extent of intraspecific variation in seed characteristics of Powell amaranth and to evaluate whether such variation was correlated with the crop rotation history of the habitat of origin. In particular, we evaluated whether seed weight, germination, and emergence patterns of Powell amaranth varied between populations originating from agricultural habitats characterized by intensive annual crop production (vegetable farms) and those originating from habitats with 3- to 5-yr periods of a perennial alfalfa crop (dairy farms with corn-alfalfa rotations). We hypothesized that (1) since Powell amaranth cannot mature successfully in established alfalfa, multiple years of perennial alfalfa on dairy farms will select for greater seed dormancy and longevity, (2) earlier spring planting dates of corn and alfalfa compared with most vegetable crops will select for earlier emergence of Powell amaranth, and (3) greater competition and lower soil moisture in the nonirrigated corn-alfalfa rotation will select for greater seed size, since larger seed size may increase the early growth rate of roots and shoots and hence their capacity to capture limited resources.

## Materials and Methods

**Seed Collection.** Seeds from 10 dairy farms and 10 vegetable farms in central New York state were collected from early September to early October in 2002 and 2003 (Table 1). Within dairy farms, fields were selected that had been in corn-alfalfa rotations for at least 10 yr with 1 to 2 yr of corn production followed by 3 to 5 yr of alfalfa. Within vegetable farms, fields were selected that had been historically under intensive annual vegetable production with at most one fallow year in the last 10 yr. The vegetable farms all produced a high diversity of crops, ensuring that in most years crops were planted later than on the dairy farms. From each site, seeds from 15 to 20 randomly selected Powell amaranth plants were

Table 1. Collection site abbreviations and nearby city in central New York state.

Type	Name	Abbreviation	Nearby city
Dairy	Autumn Ridge Farm	TH	Ithaca
	Bast Farm	BA	Spencer
	Curtis Farm	CU	Owego
	Dedrick Farm	AD	Dryden
	Efthimiou Farm	EF	Spencer
	Engelbert Farm	EN	Nichols
	H. C. Thompson Research Farm	FO	Freeville
	Hen-Lo Farm	HU	Owego
	Hoover Farm	SC	Dundee
	Myer Farm	MY	Ovid
	Seneca Valley Farm	GA	Burdett
	Smith Family Farms	SS	Clyde
	Spencer Farm	SP	Newark Valley
	Strong Farm	ST	Candor
Vegetable	Bloodnick Family Farms	BL	Apalachin
	Blue Heron Farm	BH	Lodi
	H. C. Thompson Research Farm	FS	Freeville
	Mandeville Farm	MA	Spencer
	Maple Tree Gardens	MT	Owego
	Road's End Farm	RE	Dundee
	Rose Valley Farm	RV	Rose
	Starflower Farm	LE	Candor
	Stoughton Farms	SN	Newark Valley
	Van Winkel Farm	VW	Nichols
	West Haven Farm	BS	Ithaca
	Wickham Farms	WI	Burdett

collected. Due to a lack of Powell amaranth plants in 2003 on four dairy farms (BA, CU, FO, and ST) and two vegetable farms (LE and MT) for which seeds had been collected in 2002, seeds were collected from four additional dairy farms (AD, EF, EN, SP, and SS) and two additional vegetable farms (RV and SN) in 2003. Powell amaranth was distinguished from close relatives (redroot pigweed and smooth pigweed [*A. hybridus* L.]) through close examination of floral characteristics (Costea et al. 2004). Seeds were separated from reproductive tissue using a 1.19-mm sieve followed by air column separation using a seed blower.<sup>1</sup> Separated seeds were stored dry in the dark at 5 C ( $\pm$  2 C) for 2 mo prior to germination for production of second-generation seeds.

**Greenhouse Growth.** Field-collected seeds were used to grow plants under common greenhouse conditions beginning on October 25, 2002, and November 14, 2003. These plants were used to produce a second generation of seeds to assess seed germination, emergence, and weight. Because maternal environmental conditions can have an important effect on seed characteristics of many weeds (Gutterman 2000) including Powell amaranth and its close relatives (Brainard et al. 2005a, 2005b; Kigel et al. 1977), comparison of seeds grown under common conditions is necessary to help assess genetic differences across populations.

The timing and environmental conditions associated with the storage, production, and testing of these second-generation seeds are summarized in Table 2. To avoid sampling bias based on dormancy status, 100 field collected seeds from each farm were randomly selected and set to germinate in 9-cm plastic petri dishes with 5 ml of 0.002 M gibberellic acid (GA<sub>3</sub>) in an incubator at 35 C with a 16-h day length and fluorescent lighting. This treatment resulted in greater than 95% germination of seeds from all populations (data not shown). Within 1 d of germination, 10 randomly

Table 2. Summary of timing and environmental conditions associated with seed harvest, storage, production, and testing of seeds originating from 2002 and 2003 collections.

	Environmental conditions <sup>a</sup>	Dates	
		2002	2003
Collection of seeds from farms	variable	Sep–Oct 2002	Sep–Oct 2003
Storage of field collected seed	5 C; dark	Sep–Nov 2002	Sep–Nov 2003
Dormancy breaking in growth chamber	35 C; light; 0.002 M GA <sub>3</sub>	Oct 22, 2002	Nov 11, 2003
Production of second generation seed	26/13 C; 16-h day length		
Seeding date		Oct 25, 2002	Nov 14, 2003
Dates of anthesis		Nov 30, 2002–Dec 15, 2003	Dec 17, 2003–Jan 12, 2004
Harvest dates <sup>b</sup>		Dec 31, 2002–Jan 17, 2003	Jan 21, 2004–Feb 19, 2004
Storage of second generation seed			
In glasshouse <sup>c</sup>	26/13 C; paper bags	Dec 31, 2002–Jan 31, 2003	Jan 21, 2004–Feb 26, 2004
In cooler <sup>d</sup>	5 C; dark	Jan 31, 2003	Feb 26, 2004
Growth chamber testing of germination	30/25 C; light	April 2003	Aug 2004
Seed burial in field		Nov 4, 2003	Oct 12, 2004
Exhumation of buried seed		Apr 30, 2004	Apr 28, 2005
Field monitoring of emergence	See Figure 5	May 1–Sep 10, 2004	May 1, 2005–Sep 21, 2005
Eleutriation and testing of recovered seeds			Oct–Nov 2005

<sup>a</sup> See Materials and Methods for details.

<sup>b</sup> Seeds of each plant were harvested 4 to 5 wk after anthesis.

<sup>c</sup> Seeds from all plants were stored in the glasshouse until harvest of the last plant.

<sup>d</sup> Seeds were stored in cooler until use in either growth chamber experiments.

selected seedlings from each farm were transferred to 10 2-L pots filled with a 1:1 peat moss–vermiculite mixture. Pots were placed in a greenhouse set at 26/13 C on a 16-h day length with photosynthetic photon-flux density of approximately 400  $\mu\text{mol m}^{-2} \text{s}^{-1}$  from metal halide lamps. Pots were randomly arranged within 10 blocks, each block comprised of 1 pot from each of the 20 farms. Plants were rerandomized twice weekly within blocks during the course of the experiment. Plants were watered as required and each pot was fertilized weekly with approximately 0.5 g of 15–5–15 (N–P–K) soluble fertilizer beginning 2 wk after seeding.

The timing of first appearance of floral primordia and anthesis for each individual plant was recorded. Since individual plants reached anthesis and produced mature seeds at different times, seeds from all plants were harvested from the apex (most mature seeds) of each plant 4 to 5 wk following anthesis. Seeds were stored in paper bags in this same greenhouse until final plant harvest. Seeds were then separated from reproductive tissue as described previously and transferred to a refrigerator set at 5 C until testing of germination in growth chambers or seed burial for testing of emergence (Table 2). As a result, seeds from early flowering individuals had greater storage duration at 26/23 C (greenhouse) than those maturing later. However, the timing of flowering, and hence the duration of storage did not vary significantly by habitat of origin. Within each year of the study, seeds from all populations experienced the same storage duration at 5 C (for germination testing) and the same period of burial in the soil (for emergence testing).

**Germination Testing.** In April 2003 and August 2004, following 3 (2003) or 6 (2004) mo cold storage, 50 second-generation seeds from each farm of origin were randomly selected and placed in 9-cm petri dishes with two sheets of filter paper saturated with 5 ml distilled water. Three replicates of each population were placed randomly in a single growth chamber set at 30/25 C (day/night) with a photoperiod of 16/8 h (light/dark) and photosynthetic photon-flux density of approximately 150  $\mu\text{mol m}^{-2} \text{s}^{-1}$ . Germinated seeds (radicle > 2 mm) were counted and removed daily for

10 d. Viability of second-generation seeds from each farm was tested separately by setting seeds to germinate in petri dishes as described above but with 5 ml of 0.002 M GA<sub>3</sub> and an incubation temperature of 35/30 C (day/night). As with field-collected seeds, this treatment resulted in very high germination percentages (> 97% for all populations). The viability of seeds that did not germinate following GA<sub>3</sub> treatment was tested by assessing the pressure required to crush dried seeds (Sawma and Mohler 2002). Germination and emergence data are presented as percentage of viable seeds.

**Seed Burial.** In October of 2003 and 2004 (8 to 9 mo after collection from greenhouse plants), three sets of 100 second-generation seeds from each population were sown in 10-cm-diam by 2-cm-depth PVC disks, sealed on each end with nylon mesh containing soil (coarse loamy mixed mesic Typic Fragiudept) known from eleutriation to contain no seeds of *Amaranthus* species. Disks were buried in a fallow field at the Caldwell Research Farm in Ithaca, NY, in three blocks, each containing one randomly located disk from each population. Each disk was buried horizontally at 10-cm depth within a 2-L plastic pot buried so that the soil level in the pot corresponded to the level of adjacent field soil, with approximately 1 cm of the pot rim exposed. In both years, seeds originating from all 20 collection sites from the previous fall were included. In 2004, three additional populations (MT, LE, and ST), originating from the fall of 2002 collection were also included as controls, to help distinguish effects due to year from effects due to population.

**Seed Exhumation and Emergence Monitoring.** On April 30, 2004, and April 28, 2005, disks were removed from each pot. The soil from each disk was then immediately and rapidly mixed with 80 ml of a 50:50 peat–vermiculite potting mix and spread in a 2-cm-deep layer at the top of each pot. The potting mix was added to minimize soil crusting and to simulate disturbance and fertilization that would occur during field preparation for a spring sown crop. In 2005 only, the surface soil mixture containing the Powell amaranth seeds, was separated from the rest of the soil in the pot using no-

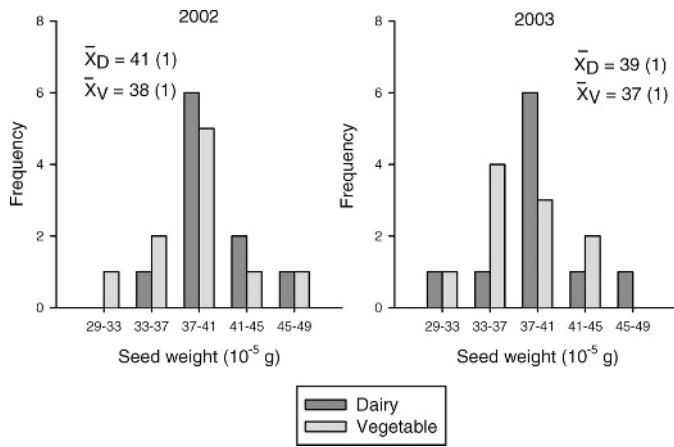


Figure 1. Histograms of Powell amaranth population mean seed dry weights by habitat of origin in 2002 and 2003. Mean values ( $\pm$  SE) by habitat of origin are also presented ( $\bar{X}_D$  = mean of 10 dairy farm populations;  $\bar{X}_V$  = mean of 10 vegetable farm populations).

secum mesh<sup>2</sup> secured to the pot with metal clips. In both years, pots were surrounded by landscaping cloth and covered with aluminum screening to prevent seed losses or gains during soil splashing from heavy rainfall events. Emergence was monitored twice weekly for the first 3 wk and then at 1- to 2-wk intervals until early September. Emerged seedlings were counted and then removed by clipping them at the soil surface.

**Seed Recovery from Soil and Viability Testing.** In 2005 only, following emergence counts, the surface layer of soil that had contained Powell amaranth seeds, was removed, dried, and the organic matter, including the seeds, was separated out using a high volume column elutriator (Gross and Renner 1989). Seeds were then sorted out by hand and counted. Seeds that crushed easily with moderate pressure were assumed to be nonviable. Hard seeds were further tested for viability via germination testing in petri dishes with 0.002 GA<sub>3</sub> at 30/25 C in light. Under these conditions, greater than 95% of the hard seed germinated for all populations.

**Statistical Analysis.** Data were analyzed using the general linear model procedures of SAS (SAS Institute 2001). Germination data were square root or log transformed as needed to improve assumptions of normality and equal variance of populations. No transformations were necessary for flowering time, emergence, or seed weight. The effects of habitat of origin (dairy vs. vegetable) on days to anthesis, seed weight, germinability, field emergence, and seed recovery following emergence (2005 only) were assessed using one-way ANOVA. The relationship between germinability in petri dishes and emergence in the field was assessed using linear regression.

## Results and Discussion

**Seed Weight.** Variation in mean dry seed weight among populations was high in both years, but did not vary significantly by habitat of origin (Figure 1). In both years, and within both habitat types, mean population seed weights ranged from about 0.30 to 0.49 mg. McWilliams (1968) found a similar range of seed weights (0.30 to 0.48 mg) for

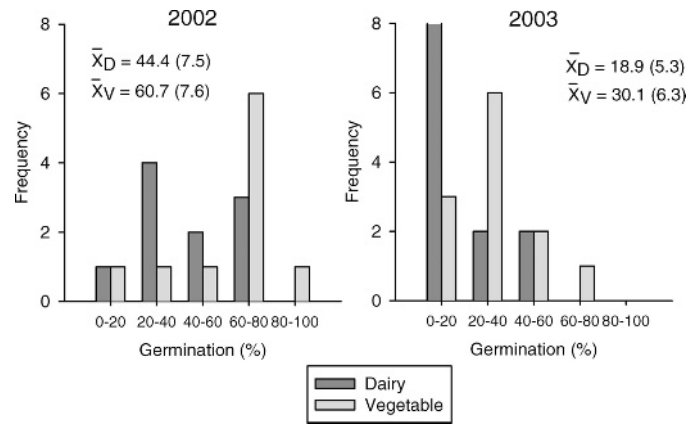


Figure 2. Histograms of Powell amaranth population mean germination percentage by habitat of origin in 2002 and 2003. Mean values ( $\pm$  SE) by habitat of origin are also presented ( $\bar{X}_D$  = mean of 10 dairy farm populations;  $\bar{X}_V$  = mean of 10 vegetable farm populations).

redroot pigweed originating from multiple latitudes following growth in a common garden in Iowa. Other studies have reported seed weights for Powell amaranth ranging from 0.41 to 0.55 mg (Weaver 1984; Senesac 1985).

Our results do not support the hypothesis of Schimpf (1977) and others (Kidson and Westoby 2000; Rees 1996; Tilman 1988) that large seed size might be an adaptive trait in habitats characterized by lower moisture or more competitive environments. Lack of differentiation across habitats in seed size may have been due to several factors. First, although the vegetable farms in this study were irrigated and the dairy farms were not, historic soil moisture levels may not have differed sufficiently across habitats to select for different seed sizes. Second, seed size may not be a heritable trait in Powell amaranth or may be correlated with other heritable traits that respond to different selective pressures. Persistent seed banks also slow the rate of evolution (Templeton and Levin 1979), so there may not have been sufficient time for microevolutionary changes to have accumulated. Finally, differences in genetic background prior to initiation of cropping practices or arrival of seeds from other habitats may obscure different evolutionary trajectories.

**Germination in Petri Dishes.** In contrast to seed weight, germination of viable seeds in petri dishes at 30/25 C was influenced by habitat of origin (Figure 2). Combined across both years of the study, the germination percentage of second-generation seeds from vegetable rotations (46%), was higher than that for dairy farms (32%) ( $P = 0.04$ ). Variation in mean population germination percentage was large; ranging from 7% (TH) to 86% (BH) for seeds originating from seeds collected in 2002, and from 3% (EN) to 75% (RV) for seeds originating from seeds collected in 2003 (Figure 2).

Frost and Cavers (1975) found a similar range of germination behavior (19 to 95%) for Powell amaranth seeds originating from six different locations but did not control for maternal environmental conditions or examine potential correlations with habitat. Our study, conducted with second-generation seeds maturing under common maternal environmental conditions suggests that some of the observed variation in germination has a genetic basis. Some of the variation in germination observed across populations may be due to variation in the timing of seed maturation, which

resulted in variation in the duration of warm storage prior to testing. However, this effect is likely to be relatively small and cannot explain differences in germination patterns between vegetable and dairy farms, since (1) seeds experienced the same duration of cold storage, which has more impact on dormancy breaking than warm storage, and (2) the average date of seed maturity was not significantly different for seeds originating from vegetable and dairy farms (data not shown).

It should be noted that germination tests of seeds in this study were conducted under a limited set of artificial conditions in a growth chamber and therefore may not reflect germination behavior in the soil. However, evidence for Powell amaranth and its close relatives from previous studies suggests that germination in light is generally well correlated with germination in darkness (Brainard et al. 2006; Senesac 1985; Weaver 1984). For example, data from Weaver (1984) shows that seed lots of redroot pigweed, Powell amaranth, and smooth pigweed with the lowest germination at 24/12 C in light were also the most dormant in darkness at 24/12 C, or in darkness or light at 35/20 C.

Lower germination among seeds originating from dairy crop rotations is consistent with the hypothesis that multiple years of perennial alfalfa on dairy farms may have selected for greater seed dormancy. This result is similar to previous work (Brainard et al. 2006) showing greater germination of Powell amaranth seeds originating on organic compared with conventional vegetable farms. The perennial alfalfa crop in dairy rotations may play a similar role as herbicides on conventional farms, reducing opportunities for seed production and hence exerting selective pressure for greater seed dormancy and longevity.

**Cumulative Field Emergence.** Field emergence was affected by habitat of origin (Figure 3). Emergence in the field following overwinter burial was higher ( $P = 0.026$ ) for seeds originating from dairy farms (62%) than for those originating from vegetable farms (52%). Farm to farm variation in emergence was large in both years and within both habitats, ranging from 9% (BH) to 75% (GA) in 2004 and from 42% (RE) to 83% (EN) in 2005 (Table 3).

The reason for overall lower seedling emergence in 2004 compared with 2005 is not clear. Seeds from populations collected in 2002 (MT, LE, and ST) that were buried as controls in both years of the emergence experiment also had lower emergence in 2004 compared to 2005 (Table 3), suggesting that year to year differences were due to climate rather than intrinsic characteristics of the seeds (although higher emergence of these populations in 2005 may also have been related to the additional year of cold storage they experienced). Higher May rainfall and temperatures in 2004 compared with 2005 (Figure 4) might have been expected to result in greater early emergence in 2004. One possibility is that warm moist conditions in spring 2004 resulted in greater incidence of fungal pathogens like *Pythium* spp. which may have killed freshly germinated seedlings prior to emergence (Davis and Renner 2005; Kremer 1993). Alternatively, differences in soil temperature, moisture, or nutrient conditions over the winter, may have resulted in different degrees of dormancy loss in the two years.

Only 1.3% of the seeds buried in the fall of 2004 were recovered from the soil and found to be viable one year later, following emergence in the summer of 2005 (Table 3). The

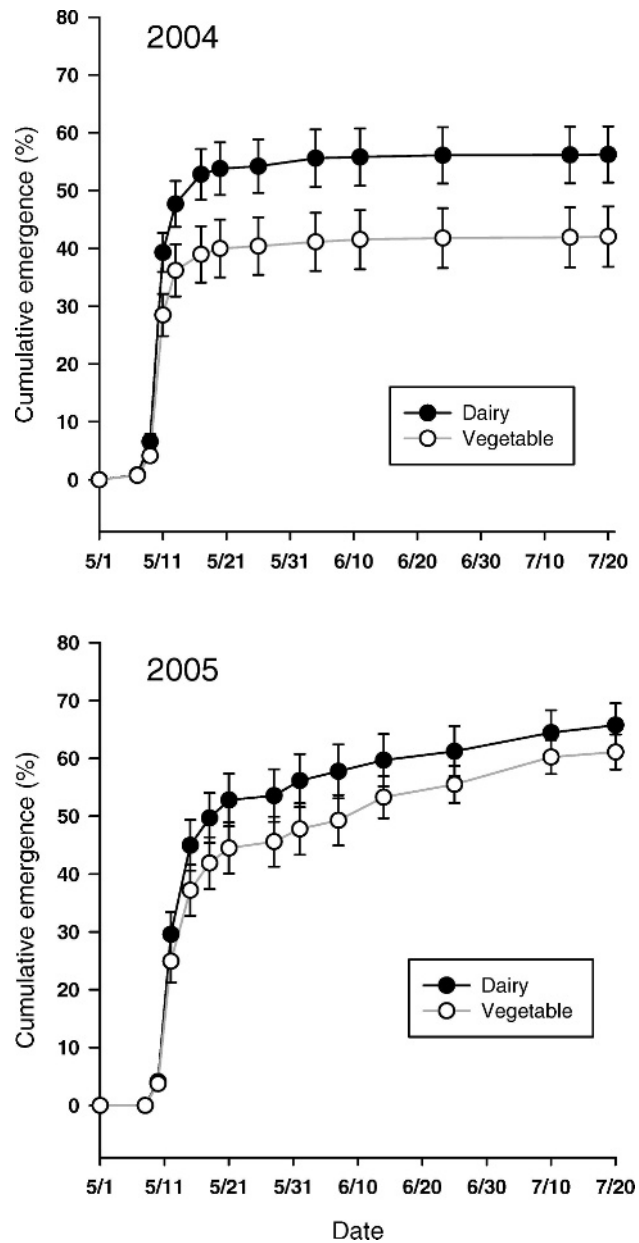


Figure 3. Mean cumulative emergence of seedlings as a percentage of seeds sown ( $\pm$  SE), by habitat of origin in 2004 and 2005.

percentage of surviving seeds ranged from 0 to 6% across populations and did not vary by habitat of origin. Since emergence and seed survival together accounted for 65% of all seeds buried in 2004, the fate of 35% of buried seeds is unknown. Possible sources of mortality for these seeds include predation, decay, or unfortunate germination. High rates of seed mortality may have been partly due to the fact that seeds were located within 3 cm of the soil surface following exhumation in the spring, and mortality of seeds is often higher in this surface zone than at greater depths (Omami et al. 1999).

The percentage of seeds emerging in this study was higher than that reported for closely related *Amaranthus* species in other studies and greater than that commonly assumed in population dynamic models. For example, summarizing several studies, Costea et al. (2004) reported that the percentage of the redroot pigweed seed bank (to 15-cm

Table 3. Mean total emergence, seed recovery, and seed mortality for 26 populations of Powell amaranth, 2004 and 2005.<sup>a</sup>

Habitat population	Total emergence		Seed recovered <sup>b</sup>		Seed mortality <sup>c</sup>			
	2004	2005	2005		2005			
	%							
Dairy	AD	NA	73.7	(2.3)	0.0	(0.0)	26.3	(0.0)
	BA	67.7 (11.1)	NA	NA	NA	NA	NA	NA
	CU	40.0 (19.5)	NA	NA	NA	NA	NA	NA
	EF	NA	59.3	(3.2)	5.7	(0.9)	35.0	(0.9)
	EN	54.7 (8.7)	82.7	(2.6)	0.0	(0.0)	17.3	(0.0)
	FO	68.7 (8.2)	NA	NA	NA	NA	NA	NA
	GA	74.7 (4.1)	65.7	(3.3)	2.0	(0.6)	32.3	(0.6)
	HU	68.0 (10.0)	58.0	(4.5)	0.7	(0.3)	41.3	(0.3)
	MY	52.3 (7.0)	82.3	(1.7)	0.7	(0.3)	17.0	(0.3)
	SC	32.0 (2.7)	44.7	(2.9)	0.0	(0.0)	55.3	(0.0)
	SP	NA	68.0	(4.0)	0.0	(0.0)	32.0	(0.0)
	SS	NA	59.3	(0.3)	0.3	(0.3)	40.4	(0.3)
	ST	37.0 (13.3)	56.0 <sup>d</sup>	(1.0)	NA	NA	NA	NA
	TH	67.3 (6.0)	70.7	(1.5)	2.0	(1.0)	27.3	(1.0)
Mean	56.2 (5.0)	66.4	(4.0)	1.1	(0.3)	32.4	(4.1)	
Vegetable	BH	9.3 (3.8)	63.3	(3.2)	0.0	(0.0)	36.7	(0.0)
	BL	30.7 (11.7)	53.3	(5.6)	NA	NA	NA	NA
	BS	41.7 (5.2)	60.0	(2.0)	3.7	(1.2)	36.3	(1.2)
	FS	41.3 (5.4)	70.7	(5.4)	1.7	(0.7)	27.6	(0.7)
	LE	42.0 (14.5)	71.8 <sup>d</sup>	(2.9)	NA	NA	NA	NA
	MA	46.7 (3.4)	76.3	(2.2)	1.3	(1.3)	22.4	(1.3)
	MT	33.3 (3.9)	72.0 <sup>d</sup>	(6.6)	NA	NA	NA	NA
	RE	54.3 (5.7)	42.0	(6.1)	0.0	(0.0)	58.0	(0.0)
	RV	NA	60.7	(0.7)	1.0	(0.6)	38.3	(0.6)
	SN	NA	60.0	(2.7)	0.3	(0.3)	39.7	(0.3)
	VW	48.3 (5.2)	63.7	(3.2)	4.7	(1.8)	31.6	(1.8)
	WI	73.0 (3.0)	67.3	(4.7)	0.0	(0.0)	32.7	(0.0)
Mean	42.1 (3.8)	61.7	(3.2)	1.4	(0.5)	35.9	(3.4)	

<sup>a</sup> Data is for seeds originally collected in 2002 or 2003 (See Table 2). Standard errors are given in parentheses.

<sup>b</sup> Viable seeds recovered via elutriation following emergence in 2005.

<sup>c</sup> Viable seeds sown (97 to 100) less emergence and seed recovery.

<sup>d</sup> Originated from 2002 collection, included as control to test environment vs. genotype effects across years.

depth) emerging each year ranges from 3 to 8%. Assuming a uniform vertical distribution of seeds in these studies and a maximum depth of emergence of 3 cm (Mohler and Galford 1997), these reported percentages would correspond to roughly 15 to 40% emergence of seeds from the germination zone (0 to 3 cm). In previous work, using seeds collected from the same set of vegetable farms, mean field emergence of five populations ranged from 24 to 52% depending on the year, habitat of origin (conventional vs. organic management practices), and rate of nitrogen fertilization (Brainard et al. 2006). Lower emergence in that study may have been due partly to the fact that seeds were stored at 5 C in a refrigerator rather than overwintering in the soil, which may have resulted in less-complete loss of dormancy. The relatively high emergence percentages observed in this trial may in part be a consequence of the maternal environment experienced by the seeds during maturation. The second-generation seeds used in this study were grown in noncompetitive greenhouse conditions under high fertility, conditions which can reduce dormancy relative to seeds growing under competition (Brainard et al. 2005a) or low fertility (Brainard et al. 2005b). However, first-generation seeds collected directly from growers' fields and buried in the fall of 2003 also had relatively high emergence rates (54%) the following spring (data not shown).

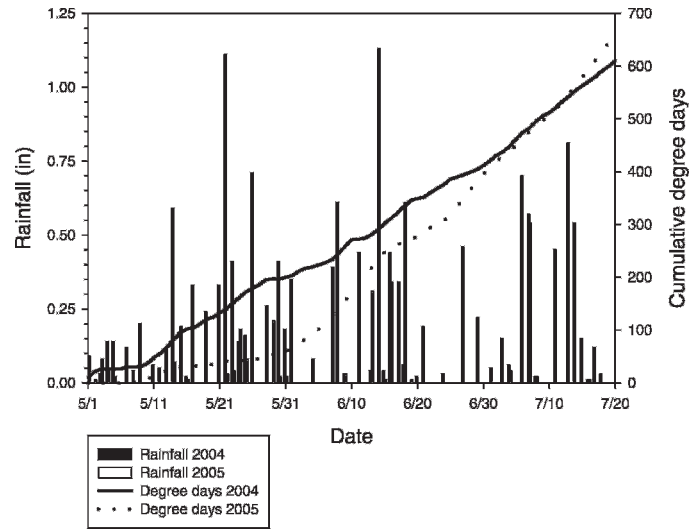


Figure 4. Rainfall (mm) and cumulative degree days (base temperature 10 C), May to July 2004 and 2005.

The higher observed emergence of seeds originating from dairy rotations is consistent with the hypothesis that such seeds have greater longevity than those originating from vegetable rotations. Since seeds from vegetable farms had higher germination in petri dishes (Figure 2), they may have experienced greater mortality due to higher unfortunate germination during the winter or early spring. This explanation is supported by the fact that germination in petri dishes was negatively correlated with emergence in the field, especially in 2004 (Figure 5). Since the fate of nonemerging seeds was not assessed in 2004, we cannot rule out the alternative possibility that seeds from vegetable farms had lower emergence due to greater dormancy in that year. However, this explanation is inconsistent with results from 2005, showing no differences in seed dormancy status (viable seeds recovered following emergence monitoring) across habitats (Table 3).

The observed higher germination and presumed greater overwinter mortality of seeds from vegetable farms compared with seeds from dairy farms may reflect divergent adaptations to crop rotation history. For the annual weed Powell amaranth, the probability of successful reproduction is higher on intensive vegetable farms than on dairy farms with a typical 3- to 5-yr crop of perennial alfalfa in their rotation. Powell amaranth seeds that germinate in alfalfa stands are unlikely to reproduce and pass their genes to successive generations. Therefore, alfalfa may exert strong selective pressure for long-lived dormant seeds. Naylor and Jana (1976) reported that dormancy was much more frequent in seeds of wild oat (*Avena fatua* L.) collected from fields with a history of frequent summer fallow than in seeds collected from fields with only occasional summer fallows. Similarly, barnyardgrass [*Echinochloa crus-galli* (L.) Beauv.] seeds collected from plants grown in continuous rice cropping systems exhibit lower dormancy than seeds from plants grown in more diverse cropping systems with both annual and perennial crops (Barrett and Wilson 1983). Likewise, higher seed germination and emergence of Powell amaranth seeds collected on organic farms vs. conventional farms may have been due to more effective herbicide-based weed management in certain crops (e.g., sweet corn) within conventional crop rotations (Brainard et al. 2006).

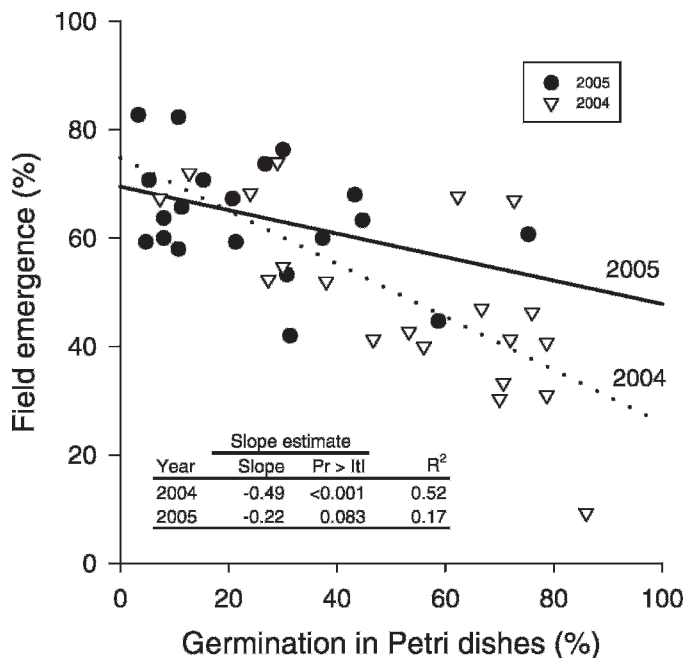


Figure 5. Relationship between seed germination in petri dishes (30/25 C) and emergence in the field following overwinter burial in 2004 and 2005.

**Emergence Rate.** The rate of seedling emergence did not vary by habitat of origin in either year (Figure 3). In 2004, over 90% of final emergence occurred within 20 d of exhumation for all 20 populations. In 2005, most emergence also occurred within 20 d of exhumation, but smaller flushes of emergence continued throughout June and July for some populations (Figures 3 and 6). Five populations showed significant delays in emergence in 2005, with between 37 and 58% of emergence occurring in June and July (Figure 6).

Rapid early emergence in 2004 was probably due in large part to above average early temperatures and rainfall in that year (Figure 4). In May 2004 rainfall and temperature exceeded long term averages by 7.5 cm and 3 C, respectively. In contrast, rainfall and temperatures in May 2005 were below long-term averages (Figure 4). High rainfall and temperatures in early June 2005 probably account for the second flush of seedling emergence observed at that time for several populations.

Our results do not support the hypothesis that Powell amaranth seeds originating from vegetable farms exhibit delayed emergence relative to those originating from dairy farms. Evolution of delayed emergence may occur in response to early weed control measures if associated increases in survival outweigh possible reductions in fecundity of survivors (Mortimer 1997). We hypothesized that later planting dates associated with many vegetable crops [e.g., tomatoes (*Lycopersicon esculentum* L.), pumpkins (*Cucurbita pepo* L.)] relative to dairy crops (corn, alfalfa), might select for later seedling emergence of weeds on vegetable farms. Early emergence of weed seedlings in vegetable cropping systems might be maladaptive, since soil preparation just prior to planting a late vegetable crop would kill early emerging weeds. The lack of differentiation in emergence timing observed in this study may be due to several factors. In addition to the explanations outlined above for lack of divergence in seed weight (e.g., possibly nonheritable trait, founder effects) two other factors should be considered. First, the diversified vegetable farms in

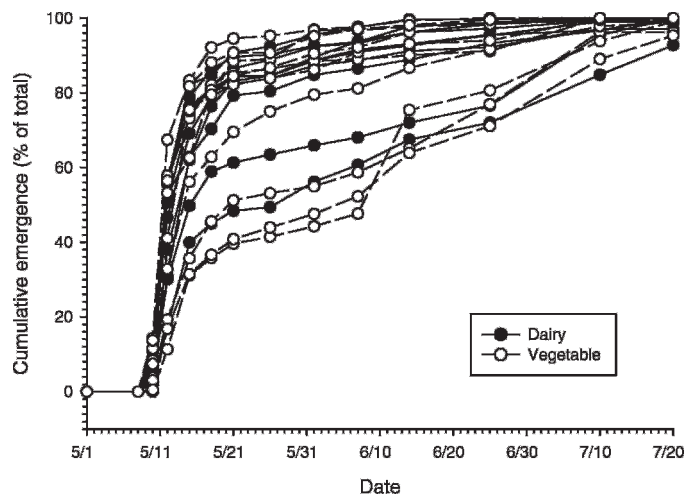


Figure 6. Mean cumulative emergence of seedlings as percentage of final cumulative emergence, by farm of origin in 2005. Habitat of origin for each farm is indicated by solid (dairy) or open (vegetable) circles.

this study have a history of both early [e.g., cabbage (*Brassica oleracea* var. *capitata*), peas (*Pisum sativum* L. ssp. *sativum*)] and late season crops in their rotations, and thus a history of potentially contradictory selection pressures operating on emergence timing. Second, the timing of emergence is tightly linked to soil disturbance which can stimulate germination of Powell amaranth through a variety of mechanisms (Costea et al. 2004). If early soil disturbance does not typically occur in fields with late sown vegetable crops, then genotypes with the potential to emerge early would remain dormant and therefore would not be exposed to selection pressure from soil disturbance during field preparation.

Lack of habitat-correlated variation in emergence timing has several important implications for weed management. For example, our results suggest that on vegetable farms with late sown crops, stale seed bed techniques have the potential to dramatically reduce Powell amaranth density. Assuming seed dormancy and soil conditions on farms were similar to those in this study, field preparation in early May, followed by herbicide application or shallow tillage in late May or early June would have reduced Powell amaranth emergence in late planted vegetable crops by over 90% on all farms in 2004 and all but a few farms in 2005 (see Figures 3 and 6). The relatively small variation in emergence timing across populations observed in this trial contrasts with previous results and suggests that simple models of Powell amaranth emergence timing may be more robust across sites than previously asserted (Brainard et al. 2006). Studies examining intraspecific variation in emergence timing of other weed species have also demonstrated relatively small intraspecific variation (Grundy 2003; Myers et al. 2004).

In summary, after controlling for maternal environmental effects, we found habitat-correlated differences in germination and emergence of Powell amaranth populations but no habitat-correlated differences in emergence-timing or seed size. Our results are consistent with the hypothesis that the presence of perennial crops in a rotation can rapidly select for seed dormancy and longevity of annual weeds. In addition, large intraspecific variation in seed size, germination, and emergence characteristics of Powell amaranth were observed. Such variation suggests that caution must be exercised in

making generalizations about species or biotypes based on one or two seed lots. For example, comparisons of fitness between resistant and susceptible biotypes of weeds like Powell amaranth based on only a few populations (e.g., Weaver and Thomas 1986) should be based on comparisons of multiple populations of each biotype (Cousens et al. 1997). Similarly, interspecific comparisons of seed germination and emergence characteristics based on one or two populations of each species (e.g., Steckel et al. 2004) cannot be reliably generalized. Finally, models of weed emergence, weed population dynamics, or weed–crop competition should benefit from increased characterization of intraspecific variation (Grundy 2003). Knowledge of the variance of key parameters is helpful for conducting sensitivity analysis and understanding the degree of uncertainty associated with model predictions.

## Sources of Materials

<sup>1</sup> South Dakota Seed Blower, Seedburo Inc., 1022 W. Jackson Boulevard, Chicago, IL 60607.

<sup>2</sup> Noseem Netting, Barre Army-Navy Store, 955 N. Main Street, Barre, VT 05641.

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## Literature Cited

- Barrett, S.C.H. and B.F. Wilson. 1983. Colonizing ability in the *Echinochloa crus-galli* complex (barnyardgrass). II. Seed biology. *Can. J. Bot.* 61:556–562.
- Baskin, C.C. and J.M. Baskin. 1998. Causes of within-species variations in seed dormancy and germination characteristics. Pages 181–237 in *Seeds: Ecology, Biogeography, and Evolution of Dormancy and Germination*. New York: Academic.
- Brainard, D.C. and R.R. Bellinder. 2001. Effect of cultivation and interseeded cover crops on weed suppression and cover crop establishment in fall kale and broccoli. *British Crop Protection Conference, Weeds—2001* 2:321–324.
- Brainard, D.C., R.R. Bellinder, and A. DiTommaso. 2005a. Effects of canopy shade on the morphology, phenology, and seed characteristics of Powell amaranth (*Amaranthus powellii*). *Weed Sci.* 53:175–186.
- Brainard, D.C., A. DiTommaso, and C.L. Mohler. 2006. Intraspecific variation in germination response to ammonium nitrate of *Amaranthus powellii* originating from organic versus conventional vegetable farms. *Weed Sci.* 54:435–442.
- Brainard, D.C., A. DiTommaso, and T.L. Setter. 2005b. Effects of maternal drought and nitrogen stress on seed germination of two populations of Powell amaranth. *Weed Sci. Soc. Am. Abstr.* 45:179.
- Bussan, A.J. and C.M. Boerboom. 2001. Modeling the integrated management of velvetleaf in a corn–soybean rotation. *Weed Sci.* 49:31–41.
- Cardina, J.E., E. Regnier, and K. Harrison. 1991. Long-term tillage effects on seed banks in three Ohio soils. *Weed Sci.* 39:186–194.
- Cardina, J., E. Regnier, and D. Sparrow. 1995. Velvetleaf (*Abutilon theophrasti*) competition and economic thresholds in conventional and no-tillage corn (*Zea mays*). *Weed Sci.* 43:81–87.
- Clements, D.R., A. DiTommaso, N. Jordan, B.D. Booth, J. Cardina, D. Doohan, C.L. Mohler, S.D. Murphy, and C.J. Swanton. 2004. Adaptability of plants invading North American cropland. *Agric. Ecosyst. Environ.* 104:379–398.
- Costea, M., S.E. Weaver, and F.J. Tardif. 2004. The biology of Canadian weeds. 130. *Amaranthus retroflexus* L., *A. powellii* S. Watson and *A. hybridus* L. (update). *Can. J. Plant Sci.* 84:631–668.
- Cousens, R., G.S. Gill, and E. Jane. 1997. Comment: number of sample populations required to determine the effects of herbicide resistance on plant growth and fitness. *Weed Res.* 37:1–4.
- Cousens, R. and M. Mortimer. 1995. *Dynamics of Weed Populations*. Cambridge, UK: Cambridge University Press.
- Davis, A.S. and K.A. Renner. 2005. Seed depth placement and soil fungal pathogens affect fatal germination of velvetleaf (*Abutilon theophrasti*). *Weed Sci. Soc. Am. Abstr.* 45:61.
- Dieleman, A., A.S. Hamill, S.F. Weise, and C.J. Swanton. 1995. Empirical models of pigweed (*Amaranthus* spp.) interference in soybean (*Glycine max*). *Weed Sci.* 43:612–618.
- Forcella, F. 1998. Real-time assessment of seed dormancy and seedling growth for weed management. *Seed Sci. Res.* 8:201–209.
- Frost, R.A. and P.B. Cavers. 1975. The ecology of pigweeds (*Amaranthus*) in Ontario. I. Interspecific and intraspecific variation in seed germination among local collections of *A. powellii* and *A. retroflexus*. *Can. J. Bot.* 53:1276–1284.
- Gross, K.L. and K.A. Renner. 1989. A new method for estimating seed numbers in the soil. *Weed Sci.* 37:836–839.
- Grundy, A.C. 2003. Predicting weed emergence: a review of approaches and future challenges. *Weed Res.* 43:1–11.
- Gutterman, Y. 2000. Maternal effects on seeds during development. Pages 59–84 in M. Fenner, ed. *Seeds: The Ecology of Regeneration in Plant Communities*. 2nd ed. New York: CABI Publishing.
- Kidson, R. and M. Westoby. 2000. Seed mass and seedling dimensions in relation to seedling establishment. *Oecologia* 125:11–17.
- Kigel, J., M. Ofir, and D. Koller. 1977. Control of the germination responses of *Amaranthus retroflexus* L. seeds by their parental photothermal environment. *J. Exp. Bot.* 28:1125–1136.
- Kremer, R.J. 1993. Management of weed seed banks with microorganisms. *Ecol. Appl.* 3:42–52.
- Linhart, Y.B. 1974. Intra-population differentiation in annual plants. 1. *Veronica peregrina* L. raised under non-competitive conditions. *Evolution* 28:232–243.
- Mayor, J.-P. and F. Dessaint. 1998. Influence of weed management strategies on soil seedbank diversity. *Weed Res.* 38:95–105.
- McWilliams, E.L., R.Q. Landers, and J.P. Mahlstede. 1968. Variation in seed weight and germination in populations of *Amaranthus retroflexus* L. *Ecology* 49:290–296.
- Menalled, F.D., K.L. Gross, and M. Hammond. 2001. Weed aboveground and seedbank community responses to agricultural management systems. *Ecol. Appl.* 11:1586–1601.
- Mohler, C.L. and B.M. Callaway. 1995. Effects of tillage and mulch on weed seed production and seed banks in sweet corn. *J. Appl. Ecol.* 32:627–639.
- Mohler, C.L. and A.E. Galford. 1997. Weed seedling emergence and seed survival: separating the effects of seed position and soil modification by tillage. *Weed Res.* 37:147–155.
- Mortimer, A.M. 1997. Phenological adaptation in weeds—an evolutionary response to the use of herbicides. *Pestic. Sci.* 51:299–304.
- Myers, M.W., W.S. Curran, M.J. VanGessel, D.D. Calvin, D.A. Mortensen, B.A. Majek, H.D. Karsten, and G.W. Roth. 2004. Predicting weed emergence for eight annual species in the northeastern United States. *Weed Sci.* 52:913–919.
- Naylor, J.M. and S. Jana. 1976. Genetic adaptation for seed dormancy in *Avena fatua*. *Can. J. Bot.* 54:306–312.
- Omami, E.N., A.M. Haigh, R.W. Medd, and H.I. Nicol. 1999. Changes in germinability, dormancy and viability of *Amaranthus retroflexus* as affected by depth and duration of burial. *Weed Res.* 39:345–354.
- Rees, M. 1996. Evolutionary ecology of seed dormancy and seed size. *Phil. Trans. R. Soc. Lond. B* 351:1299–1308.
- Rice, K.J. and N.C. Emery. 2003. Managing microevolution: restoration in the face of global change. *Front. Ecol. Environ.* 1:469–478.
- Salisbury, E.J. 1942. *The Reproductive Capacity of Plants*. London: G. Bell and Sons.
- SAS Institute. 2001. *SAS/STAT User's Guide Version 8-1*. Cary, NC: SAS Institute. 1030 p.
- Sawma, J.T. and C.L. Mohler. 2002. Evaluating seed viability by an unimbibed seed crush test with comparison to the tetrazolium test. *Weed Technol.* 16:781–786.
- Schimpf, D.J. 1977. Seed weight of *Amaranthus retroflexus* in relation to moisture and length of growing season. *Ecology* 58:450–453.
- Schweizer, E.E. and R.L. Zimdahl. 1984. *Amaranthus powellii* interference in sugarbeet (*Beta vulgaris*). *Weed Sci.* 33:518–520.

- Senesac, A.F. 1985. Aspects of the biology and control of pigweed (*Amaranthus* spp.) in New York. Ph.D. dissertation. Ithaca, NY: Cornell University.
- Sosnoskie, L.M., C.P. Herms, and J. Cardina. 2006. Weed seedbank community composition in a 35-yr-old tillage and rotation experiment. *Weed Sci.* 54:263–273.
- Steckel, L.E., C.L. Sprague, E.W. Stoller, and L.M. Wax. 2004. Temperature effects on germination of nine *Amaranthus* species. *Weed Sci.* 52:217–221.
- Swinton, S.M. and R.P. King. 1994. A bioeconomic model for weed management in corn and soybean. *Agric. Syst.* 44:313–335.
- Tardif, F.J., I. Rajcan, and M. Costea. 2006. A mutation in the herbicide target site acetohydroxyacid synthase produces morphological and structural alterations and reduces fitness in *Amaranthus powellii*. *New Phytol.* 169:251–264.
- Templeton, A.R. and D.A. Levin. 1979. Evolutionary consequences of seed pools. *Am. Nat.* 114:232–249.
- Tilman, D. 1988. *Plant Strategies and the Dynamics and Structure of Plant Communities*. Monographs in Population Biology 26. Princeton, N.J.: Princeton University Press.
- Weaver, S.E. 1984. Differential growth and competitive ability of *Amaranthus retroflexus*, *A. powellii* and *A. hybridus*. *Can. J. Plant Sci.* 64:715–724.
- Weaver, S.E. and A.G. Thomas. 1986. Germination responses to temperature of atrazine-resistant and -susceptible biotypes of two pigweed (*Amaranthus*) species. *Weed Sci.* 34:865–870.

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