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DIRECT MEASUREMENT OF THE SEED BANK AGE STRUCTURE OF A SONORAN DESERT ANNUAL PLANT

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Abstract. We describe a new approach to determining the age structure of seed banks of natural plant populations and apply it to a natural population of the Sonoran Desert winter annual, *Pectocarya recurvata* (Boraginaceae). Unlike other ^{14}C techniques, tandem accelerator mass spectrometry (TAMS) counts the number of carbon isotope atoms, permitting high precision with small samples. Aboveground nuclear bomb tests caused atmospheric ^{14}C levels to peak in 1963. Their subsequent gradual decline provides a signal for aging seed banks with TAMS. We constructed a calibration curve using seeds with known dates of production during 1980–1995, then used it to age 53 seeds sampled from a natural seed bank in 1993, at the Desert Laboratory in Tucson, Arizona. Seed number declined with age at an approximately exponential rate, with the oldest recovered seed having an estimated age of 5 yr (95% CI = ±2.3 yr). The seed bank age structure was judged more than adequate to buffer this population from typical fluctuations, based on an examination of 15 yr of population dynamic data. The TAMS technique has strong potential for answering a broad range of ecological and evolutionary questions requiring post-1963 age determinations and for which a several-year confidence interval is acceptable.

Key words: age determination; bet hedging; ^{14}C; desert annual; nuclear bombs; Pectocarya recurvata*; population persistence; seed bank age structure; seed dormancy; Sonoran Desert; tandem accelerator mass spectrometry (TAMS); viable seed number vs. seed age.*

INTRODUCTION

Not all viable seeds produced in natural plant populations germinate in the season following their production. If a portion survive to subsequent years, the result is an age-structured seed bank. The seed bank age structure is a critical determinant of many aspects of basic and applied population and community ecology, as well as of evolutionary dynamics (Leck et al. 1989). What are extinction probabilities and effective population sizes? How do life history attributes respond to natural selection and determine population growth rates? What communities will return after human or natural disturbances of various durations? Which weed control strategies are likely to work? The answers to these and other questions about metapopulation dynamics, bet-hedging adaptations, community resilience, successional dynamics, and species coexistence all require the measurement and understanding of seed bank dynamics.

Desert annuals provide an important nexus for consideration of many of the ecological and evolutionary questions regarding seed banks. They are one of the classic examples of plants assumed to depend on seed bank carryover (Juhren et al. 1956), yet scant data on actual seed age structures is available. Seed banks are posited to be critical to desert annual populations persisting in variable and unpredictable environments (Cohen 1966, Pake and Venable 1996). Most evolutionary theories of bet hedging as an adaptive response to unpredictably variable environments have focused on desert annual seed banks as model systems (Philippi and Seger 1989). Also, desert annual seed banks have played an important role in developing theoretical ideas about the population dynamic functions of dispersal and variance-mediated species coexistence (Cohen and Levin 1985, Brown and Venable 1986, Chesson and Huntley 1989).

Since seeds are virtually inert metabolically, there are no structural indices of age, analogous to the tree rings or tuber rings, which permit age estimates for the

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growth stage of some plants. Thus, while delayed germination is a critical aspect of many ecological and evolutionary questions, determining the age structure of seed banks in natural populations has remained a recalcitrant problem. The most successful approach has been the cumbersome practice of following seed cohorts for many years or even decades (e.g., the Beal experiment—Darlington [1931], [1951], Darlington and Steinbauer [1961]; the Duvel experiment—Duvel [1902], Goss [1924], Toole and Brown [1946]; Roberts and Feast 1973, Smith 1983, Kalisz 1991).

Tandem accelerator mass spectrometry (TAMS), together with mid-20th century political history, provides a new method to determine seed bank age structure. $^{14}C/^{13}C$ fractions of samples as small as 100 μg of carbon are compared to standards using tandem accelerators (Donahue et al. 1990). Unlike other carbon-dating methods that monitor scintillations produced by radioactive decay, the TAMS method counts the actual number of carbon isotope atoms in a sample. This permits high precision and allows the use of very small samples, like seeds. Atmospheric levels of ^{14}C peaked in 1963, a time of extensive aboveground nuclear bomb tests, and have been gradually declining since (T. Lange, *unpublished data).* By comparing seeds of unknown age sampled from the soil seed bank to the $^{14}C/^{13}C$ signatures of seeds of known age, seed bank age structure can be reconstructed. This snapshot of age structure is not subject to confounding by habitat modification, as typically occurs with other techniques (e.g., burial in a jar or suppressing new seed production). It has the additional advantage of being able to age old seeds as readily as young ones, since we do not have to wait for seeds to die.

In this paper we ask, how accurate is TAMS for dating seeds of known age, and can it be used to reconstruct the seed bank age structure of a Sonoran Desert annual plant? We chose *Pectocarya recurvata,* because it is a common winter annual with medium seed size, for which we have long-term data on field germination and population sizes (Venable and Pake 1999). Thus the results are relevant to ongoing research on population and community dynamics of desert annuals. Specifically, we ask the following questions: Does *Pectocarya recurvata* have a between-year seed bank? And, what are the mean and maximum ages of seeds in a random sample of its seed bank?

Methods

Calibration

To determine the age of a natural seed bank, we first constructed a calibration curve with seeds of known age. We have collected fresh seeds of *Pectocarya recurvata* Jtn. (Boraginaceae; mean seed mass, ~0.95 mg) [hereafter referred to as *Pectocarya*] and *Plantago insularis* (Plantaginaceae; mean seed mass, ~0.95 mg) [hereafter referred to as *Plantago*], and we have stored them in the laboratory since 1982, as part of a long-term research project on desert annuals (Venable and Pake 1999). These seeds of know age were all collected at maturity from parent plants at the Desert Laboratory in Tucson, Arizona, USA. To calibrate the $^{14}C/^{13}C$ readings from tandem accelerator mass spectrometry (TAMS), we used samples from all of our *Pectocarya* accessions with known age (21 samples, 1988–1995) supplemented for the earlier years with seeds of *Plantago* (7 samples, 1983–1988). All of the *Plantago* and three of the *Pectocarya* samples were bulk samples (two or three seeds per sample for *Pectocarya*; three to five seeds per sample for *Plantago,* yielding ≥350 μg C upon combustion). Eighteen of the *Pectocarya* samples consisted of individual seeds. $^{14}C/^{13}C$ readings for 35 previously analyzed bulk samples of cotton seeds (from 1980–1992) were also used to construct the calibration curve.

Each *Plantago* and *Pectocarya* sample was pretreated, combusted, and run through the tandem accelerator at the Accelerator Mass Spectrometer Laboratory (AMS) at the University of Arizona. Pretreatment removes organic contaminants that may be present on the seed. Each individual seed was subjected to 2 h of diluted NaOH solution, distilled H_2O rinse, 2 h of diluted HCl solution, distilled H_2O rinse, 2 h of diluted NaOH, and a final distilled H_2O rinse. Then the seeds were dried overnight in a drying oven. The dried seeds were added into a combustion tube with copper oxide (CuO = [sample mass/2] × 100; masses measured in milligrams), where they were reduced to CO_2 in the vacuumed combustion setup at the AMS facility. Following purification, each sample was given a $\delta^{13}C$ test to determine the $^{13}C/^{12}C$ ratio. This was done to make sure that any fractionation of the different isotopes of carbon did not occur during the combusting process. The carbon dioxide was then converted to carbon monoxide and fixed to iron (Fe) to be made into graphite.

The TAMS instrument operates in the following manner: the graphite samples are loaded into a wheel, which is inserted into a cesium sputter-type ion source on the accelerator. The cesium ions interact with the graphite and eject the carbon ions. Negatively charged carbon ions are accelerated and progressively purified through the tandem electrostatic accelerator. The accelerator has two Faraday cups (one low energy and one high energy), which are used to quantify the isotopes of carbon. This instrument is described in detail in Donahue (1995).

The AMS Laboratory at the University of Arizona is one of four such laboratories in the USA that perform radionuclide measurements on small samples. TAMS

facilities are also available in many other developed countries. Prices are currently comparable to those for the older ^{14}C-counter technology. The University of Arizona laboratory routinely measures >5000 ^{14}C samples/yr.

The ^{14}C signal that is used to age seeds is called fraction modern (f-modern), and it is calculated using the following expression:

$$\text{f-modern} = \frac{(^{14}C/^{13}C)_s}{(^{14}C/^{13}C)_{1950}}$$

where $(^{14}C/^{13}C)_s$ represents the carbon ratio of the sample and $(^{14}C/^{13}C)_{1950}$ represents the carbon ratio of standard samples from 1950 (Donahue et al. 1990).

In the course of this investigation, we noticed that samples yielding <350 μg C tended to have lower f-moderns than larger samples of the same age. Therefore, a mass correction curve was constructed by comparing f-modern values of small mass seeds (100–600 μg) of known ages with f-modern values of large samples (≥1.0 mg) of seeds of the same known age. A least-squares linear regression of f-modern vs. age was performed on 47 large samples (≥1.0 mg) of known age (including the cotton data). Fractions moderns for an additional 15 small samples (≤600 μg) of known age tended to plot below this regression line (especially the smaller of these). Thus, the fraction modern for each of the small and large samples was subtracted from the predicted f-modern for seeds of the same age from the large-sample regression. These differences were regressed against the natural log of amount carbon (*M*, measured in micrograms) for each sample, yielding the following formula for mass correction (MC): MC = 0.139 − 0.0212 × ln(*M*). Mass corrections calculated in this way were added to the measured f-modern for seeds of mass ≤600 μg in subsequent analyses.

A curve to estimate age from f-modern was obtained by a least-squares linear regression of f-moderns on year of seed production (1980–1995) using our *Pectocarya* and *Plantago* seeds, which were mass corrected when mass ≤600 μg, and the bulk samples of cotton seeds. Altogether, there were 63 values for 14 yr of the 16-yr time span (mean no. readings/yr > 4). Terms for species and for species × year interaction were included in the model to test for differences among *Pectocarya*, *Plantago*, and cotton (using the INSIGHT procedure of SAS). As there were no significant differences among the three species in slope or intercept, the species and species × year terms were dropped from the final model (Fig. 1).

The year of seed production (*x*-axis) can be predicted from f-modern (*y*-axis) by inverting this simple regression equation (i.e., by solving the equation for *x* in terms of *y*: year = −(f-modern − 26.002)/0.0125).

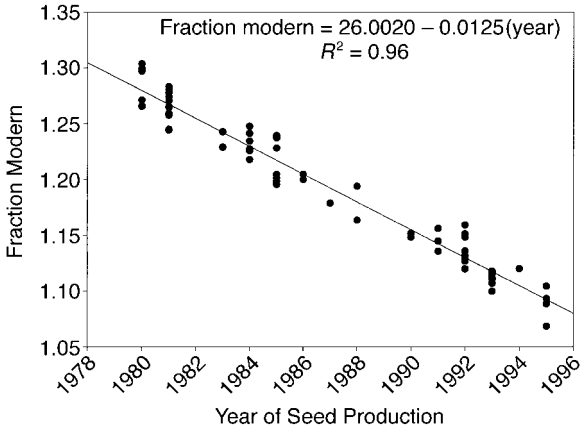

FIG. 1. Calibration curve relating the $^{14}C/^{13}C$ signal (fraction modern) to year. The regression was constructed from results of tandem accelerator mass spectrometry (TAMS) runs using 21 samples of *Pectocarya recurvata* seeds, seven samples of *Plantago insularis* seeds, and 35 samples of cotton seeds, all with known year of seed production.

The 95% confidence intervals for the estimated years of seed production were calculated with the CI formula for inverse predictions from linear regressions (Zar 1984:276, Sokal and Rohlf 1995:492).

Aging unknown seeds

To determine the age structure of the seed bank of a natural population of *Pectocarya*, we analyzed 177 soil samples (22.9 cm^2 area, 2.5 cm deep), which were collected during February 1993 along a transect of permanent demographic plots using a stratified random sampling technique (Pake and Venable 1996). Virtually all viable winter annual seeds at this site are found in the upper 2.5 cm of the soil, which typically has a hard crust (Pake and Venable 1996). Half of the samples were collected under shrubs (mostly *Larrea tridentata*) and half in the open, since vegetation cover was ~50% (see Pake and Venable [1995] for description of the site and long-term plots). Only 12 of the 53 recovered seeds that were analyzed came from under shrubs. There was no significant difference in the predicted age structure of open vs. under-shrub habitats (but statistical power to test for a difference was low). Thus, the results for shrub and open samples were pooled for subsequent analysis.

Samples were air dried in the laboratory and separated into fractions with several U.S.A. Standard Testing Sieves (Tyler, Mentor, Ohio, USA). All fractions that potentially contained *Pectocarya* seeds were examined for intact *Pectocarya* seeds, a small amount at a time, under a dissecting microscope. The spiny recurved seeds (technically nutlets) of *P. recurvata* are easily distinguished morphologically from those of *P.*

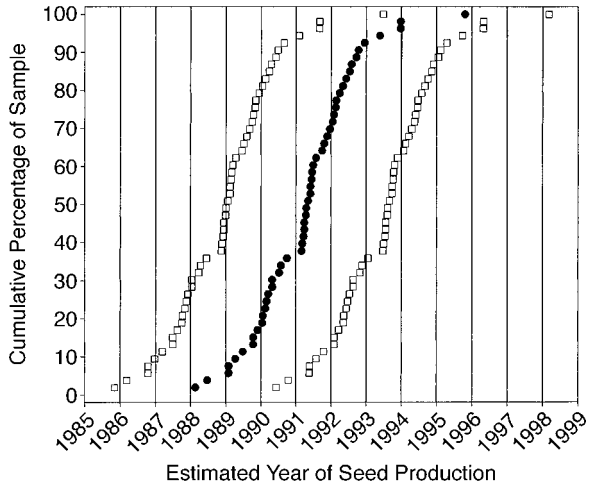

FIG. 2. Cumulative frequency distribution of the estimated year of seed production for 53 seeds sampled in February 1993, from the seed bank of a natural population of *Pectocarya recurvata.* The middle point (circle) is the estimated date of origin for each seed. The right and left points (squares) delimit the 95% CI on each estimate.

heterocarpa, *P. platycarpa*, and *Lapula rezdowski*, which also occur at low frequency in the sampled area. These are the only other winter annuals at this site with spiny nutlets that are somewhat similar to those of *P. recurvata.*

At the time of the combustion treatment, each *Pectocarya* ''seed'' was cut or poked through the seed coat to determine viability. Seeds with light-colored, fleshy, or oily endosperm were regarded as viable. This technique is more reliable than tetrazolium chloride for this species, because some deeply dormant viable seeds show minimal staining with tetrazolium (Pake and Venable 1995). Yet, embryos and endosperm in the seed bank under natural field conditions undergo easily recognized changes upon death (desiccation, decay).

By February, the germination season for *Pectocarya* is complete at this site (it has been bounded by October and January for the last 15 yr), but new seeds on growing plants have not yet dropped (seed set occurs in March and April). Thus, all sampled seeds were produced during or prior to Spring 1992 and had the opportunity to germinate during at least one germination season (October 1992–January 1993). Since this germination season was quite wet, all seeds that we tested had experienced at least one good germination season without germinating.

Of the 149 recovered viable *Pectocarya* seeds of unknown age, 63 were randomly selected for aging. Mass-corrected f-modern values were determined for the seeds. The readings for 10 seeds were discarded, due to bad run days at the AMS Laboratory (e.g., when it was decided that electrical currents had not been strong enough to produce reliable results for a particular day of operation). The inverse of the previously constructed calibration curve and the corresponding confidence interval formula for inverse prediction were used to estimate the year each seed was produced and its 95% CI.

RESULTS

An important result for evaluating the accuracy of tandem accelerator mass spectrometry (TAMS), and its utility for aging seed banks, is the scatter around the regression curve in Fig. 2. The scatter for the regression of f-modern on year is quite small ($R^2 = 0.96$, $P < 0.0001$), indicating that TAMS successfully recovers f-modern values that are strongly determined by the date of seed production. The pertinent question for dating seeds of unknown age is as follows: How does this curve and its scatter translate into confidence limits on the year of seed production for a seed with known f-modern? These 95% confidence intervals were determined to be $\sim \pm 2.3$ yr (differing slightly for each seed). Thus, while we cannot unequivocally determine the exact year of seed production, we can place confidence bounds around the estimated dates and thereby test specific hypotheses, such as no seed bank, or a specified maximum age of seeds.

When the prediction curve was applied to the 53 viable seeds from the 1993 seed bank of *Pectocarya*, estimated dates of production ranged from Spring 1988 to Spring 1996 (Fig. 2). The percentage of viable seeds estimated to be 5-yr-old was 3.8%; 4-yr-old, 7.5%; 3-

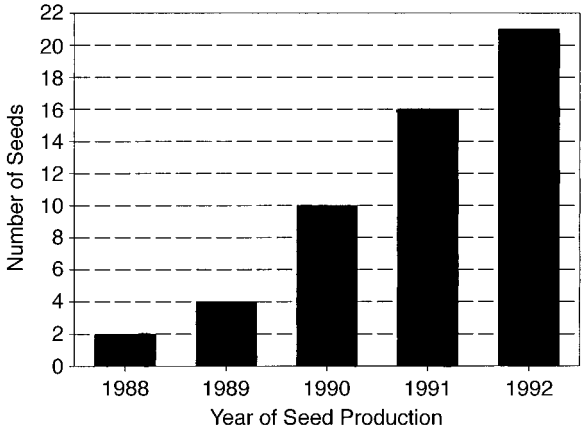

FIG. 3. Frequency histogram of estimated dates of seed production for 53 seeds sampled in February 1993, from the seed bank of a natural population of *Pectocarya recurvata.* Seeds with estimated dates of production after spring 1992 (see Fig. 2) were combined with those estimated to have been produced during spring 1992, because this was the last time seeds were produced prior to the sampling date.

yr-old, 18.9%; 2-yr-old, 30.2%; and 1-yr-old, 22.6%. An additional nine seeds were estimated to come from after Spring 1992, but this is biologically impossible, since no new seeds were produced between Spring 1992 and the time of seed bank sampling in February 1993. However, only one of these ''post-1992'' seeds had a 95% CI ($\pm$2.3 yr) that did not include Spring 1992 (Fig. 2). This is to be expected with 53 seeds, since five percent of samples are likely to have true dates of production outside of 95% CIs. When these post-1992 seeds are grouped with the Spring 1992 seeds, 39.6% of the sample were in the youngest one-year-old age category (Fig. 3).

Six seeds had confidence limits prior to and not including 1992 and thus, with the specified level of confidence, have remained dormant and viable through at least two germination seasons (fall 1991 and fall 1992). We can also conclude that few if any of the sampled seeds were produced prior to 1986 (i.e., remained viable but dormant through greater than seven germination seasons). The median seed was produced in 1991 and was 2-yr-old at the time of sampling. Together, these results suggest that, in February 1993, *Pectocarya* had a fairly well-developed seed bank age structure, with seed ages skewed toward young dates (Fig. 3).

Discussion

An ideal seed bank aging technique would have 95% CI $<$ 0.5 yr, so as to unequivocally identify the year of production of most seeds. At present, no such technique is available. Nevertheless, the TAMS technique reported here, which generated confidence limits of $\pm$2.3 yr, can provide results that are useful for making strong inferences regarding many ecological and evolutionary consequences of seed banks: Is there an age-structured seed bank? What is its general shape? What is the 95% confidence limit on the oldest sampled seed? All of these questions can be answered from a single sample collected and evaluated in a single year, as opposed to previous longitudinal techniques which require following a seed cohort for years, often under somewhat artificial conditions.

A potential shortcoming of the TAMS technique is it's inability to distinguish seed production dates before 1963 from those since 1963. Yet, for most species, only a small fraction, if any, of the seeds in a natural seed bank will be $>$37-yr-old, and the possibility of their existence will be clearly evident from the distribution of estimated seed ages (e.g., the seed age distribution of *Pectocarya* reported here tails off long before 37 yr). Determining the ages of such old seeds is even more difficult using longitudinal studies ($>$40 yr experiments).

Individual *Pectocarya* seeds are approaching the current size limit for this approach, forcing us to apply a mass correction to individual seed samples. While our 0.95-mg seeds are smaller than the typical herb seed (Silvertown and Lovett-Doust 1993:164, Table 10.1), there are many species with considerably smaller seeds, so other approaches will have to be taken to age their seed banks. One approach would be to obtain mean readings for groups of seeds and attempt to reconstruct the variance in age of individual seeds from the variance of the means of a known number of seeds. We conclude that, while TAMS falls short of being the ideal technique, it represents an important advance in the rapid aging of seed banks.

The utility of the technique is illustrated by its application to *Pectocarya.* A null hypothesis of no seed bank carryover must be rejected because (1) at least some seeds did not germinate in the good germination season of the fall of 1992 and were still viable at the time of sample collection, and (2) six of these viable seeds had confidence limits prior to and not including 1992 and thus, with the specified level of confidence, had remained dormant and viable through at least two germination seasons. We were also able to place a 7-yr bound on the maximum age of sampled seeds (few if any of the sampled seeds were produced before 1986). Thus, the results indicate a moderately developed seed bank age structure, though almost seeds are not very old (median equals 2-yr-old).

These results have some interesting implications for the biology of *Pectocarya* and, by extension, desert annuals in general. It may be initially surprising that the seed bank of a desert annual is not older. Yet, adequate precipitation for substantial germination of nondormant seeds has existed at the Desert Laboratory in 10 of the last 15 yr (D. L. Venable, *unpublished data*). Our study site is located on the eastern edge of the Sonoran Desert, where precipitation is fairly high (mean, 250–300 mm/yr). We would expect *Pectocarya* to have an older aged seed bank in sites with a lower frequency of years with good germination season rains, such as sites in the heart of the Sonoran Desert near the delta of the Colorado River or in Death Valley. We would also predict that some other species in the same community at the Desert Laboratory would have older aged seed banks. Field experiments comparing the density of germinated seedlings with the density of post-germination-season viable seeds indicate that *Pectocarya* has a relatively high germination fraction, compared to other species in this community (Pake and Venable 1996, Venable and Pake 1999). Another factor contributing to the young ages of seeds is that seed bank densities in this community were low following the extended late 1980s drought, and the 1993 census was conducted following several years of population increase (Pake and Venable 1996, Venable and Pake

1999). Age structures are skewed toward the younger age classes during such times of population increase.

Even this relatively young seed bank should permit the *Pectocarya* population at the Desert Laboratory to withstand several consecutive years of good conditions for germination, coupled with total reproductive failure. During the last 15 yr, there has never been more than one consecutive year of total reproductive failure, or three consecutive years in which conditions for germination were good and the product of survival of germinating seeds to reproduction times mean fecundity was less than one (Venable and Pake 1999). While a very long-term data set on population dynamics would be necessary to capture unusual extreme events, this 15-yr data set suggests that the observed seed bank age structure of *Pectocarya* is more than adequate to buffer the population from typical fluctuations.

From this investigation we conclude that determining seed bank age structure with TAMS is feasible and has strong potential for answering a broad range of basic and applied ecological and evolutionary questions. The theoretical basis of this technique suggests that other species should follow the same calibration curve that we estimated. This was confirmed in this study by the lack of an empirical difference between cotton, *Plantago*, and *Pectocarya* seeds of known ages. Thus, it seems reasonable to apply our calibration curve to other species for which an assortment of reference seeds of known age are not available. Also, this technique should be directly extendable to the aging of other small recent biological structures, such as fish eggs or diapausing crustaceans.

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Literature Cited

Brown, J. S., and D. L. Venable. 1986. Evolutionary ecology of seed-bank annuals in temporally varying environments. American Naturalist **127**:31–47.

Chesson, P. L., and N. Huntley. 1989. Short-term instabilities and long-term community dynamics. Trends in Ecology and Evolution **4**:293–298.

Cohen, D. 1966. Optimizing reproduction in a randomly varying environment. Journal of Theoretical Biology **12**: 119–129.

Cohen, D., and S. A. Levin. 1985. The interaction between dispersal and dormancy strategies in varying and heterogeneous environments. Pages 110–122 *in* E. Teromato and M. Yomaguti, editors. Mathematical topics in population biology, morphogenesis and neurosciences. Proceedings of International Symposium, Kyoto, Japan.

Darlington, H. T. 1931. The fifty-year period for Dr. Beal's seed viability experiment. American Journal of Botany **18**: 262–265.

Darlington, H.T. 1951. The seventy-year period for Dr. W. J. Beal's seed viability experiment. American Journal of Botany **38**:379–381.

Darlington, H. T., and G. P. Steinbauer. 1961. The eighty-year period for Dr. Beal's seed viability experiment. American Journal of Botany **48**:321–325.

Donahue, D. J. 1995. Radiocarbon analysis by accelerator mass spectrometry. International Journal of Mass Spectrometry and Ion Process **143**:235–245.

Donahue, D. J., T. W. Linick, and A. J. T. Jull. 1990. Isotoperatio and background corrections for accelerator mass spectrometry radiocarbon measurements. Radiocarbon **32**:135–142.

Duvel, J. W. T. 1902. Seeds buried in soil. Science **17**:872–873.

Goss, W. L. 1924. The vitality of buried weed seeds. Journal of Agricultural Research **29**:349–362.

Juhren, M., F. W. Went, and E. Phillips. 1956. Ecology of desert plants. IV. Combined field and laboratory work on germination of annuals in the Joshua Tree National Monument, California. Ecology **37**:318–330.

Kalisz, S. 1991. Experimental determination of seed bank age structure in the winter annual *Collinsia verna.* Ecology **72**:575–585.

Leck, M. A., V. T. Parker, and R. L. Simpson. 1989. The ecology of soil seed banks. Academic Press, San Diego, California, USA.

Pake, C., and D. L. Venable. 1995. Is coexistence in Sonoran Desert annuals mediated by temporal variability in reproductive success? Ecology **76**:246–261.

Pake, C. E., and D. L. Venable. 1996. Seed banks in desert annuals: implications for persistence and coexistence in a variable environment. Ecology **77**:1427–1435.

Philippi, T., and J. Seger. 1989. Hedging one's evolutionary bets, revisited. Trends in Ecology and Evolution **4**:41–44.

Roberts, H. A., and P. M. Feast. 1973. Emergence and longevity of seeds of annual weeds in cultivated and undisturbed soil. Journal of Applied Ecology **10**:133–143.

Silvertown, J. W., and J. Lovett-Doust. 1993. Introduction to plant population biology. Blackwell Science, Oxford, UK.

Smith, B. H. 1983. Demography of *Floerkea proserpinacoides,* a forest-floor annual. III. Dynamics of seed and seedling populations. Journal of Ecology **71**:413–425.

Sokal, R. R., and F. J. Rohlf. 1995. Biometry. Freeman, New York, New York, USA.

Toole, E. H., and E. Brown. 1946. Final results of the Duvel buried seed experiment. Journal of Agricultural Research **72**:201–210.

Venable, D. L., and C. E. Pake. 1999. Population ecology of Sonoran Desert annual plants. Pages 115–142 *in* R. H. Robichaux, editor. The ecology of Sonoran Desert plants and plant communities. University of Arizona Press, Tucson, Arizona, USA.

Zar, J. H. 1984. Biostatistical analysis. Prentice Hall, Englewood Cliffs, New Jersey, USA.