Weed Science. 1993. Volume 41:185–193

The Germination Stimulant AC94377 Reduces Seed Survival of Wild Mustard (*Sinapis arvensis*)[1]

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Abstract. **Annual application of AC-94377 at 3.4 kg ha^{-1} in the field reduced survival of shallowly buried (1.25 cm deep), undisturbed wild mustard seed compared to untreated check seed in two 4-yr-long trials. By the second fall, greater than twofold more untreated check seed survived as did AC-94377-treated seed. Moreover, no AC-94377-treated seed survived beyond year three following treatment in fall alone or fall plus spring in each of 3 yr. In contrast, 25% of untreated check seed survived into the fall of year four. AC-94377 applied in spring alone, fall alone, or both spring and fall for each of 4 yr progressively reduced seed survival. Seed survival expressed as a percent of the initial number of seed buried was best modeled as a negative exponential function of time in years. In the greenhouse, more wild mustard seed on the soil surface established after AC-94377 treatment at 3.4 kg ha^{-1} when enclosed in large seed packets (5 by 12 cm), like those used in the field, than when in small seed packets (5 by 6.25 cm), whether or not the packets contained soil. When soil was added to either sized seed packet, fewer seed survived compared to seed not in seed packets or seed in packets without soil. Thus, it is likely that the field seed survival study underestimated effectiveness of AC-94377 to reduce wild mustard seed survival. Nomenclature: AC-94377, 1-(3-chlorophthalimido)cyclohexanecarboxamide; wild mustard, *Sinapis arvensis* #[3] SINAR.**

Additional index words: **Germination stimulation, phthalimides, seed persistence, SINAR.**

INTRODUCTION

Annual weeds are recurring problems on farmland because of seed dormancy and the buildup of persistent seed populations in the soil (21). Weed seed in the soil seedbank often germinate under favorable environmental conditions over several subsequent growing seasons. Chemicals that break weed seed dormancy and stimulate premature germination have been studied (26) in the hope that they can be used to speed the loss of weed seed from the soil seedbank. Emerged weeds can then be killed by herbicides, tillage, or other means. Germinated weed seed that cannot emerge because of physical barriers, such as crusting or depth, are subject to decay by microorganisms.

Chemical stimulation of dormant weed seed germination by nitrogen fertilizers, herbicides, and either natural or synthetic plant growth regulators was reviewed (26). The N-substituted phthalimide AC-94377 enhanced dark germination of dormant seed of numerous weeds (4, 8, 19) both in petri dishes (4, 19) and soil (4, 8) in the greenhouse. These studies demonstrated that AC-94377 did not kill dormant seed. AC-94377 has gibberellin-like activity in a range of gibberellin bioassays (24) and competitively inhibited GA_4 binding in cucumber (*Cucumis sativus* L.) hypocotyl extracts (29). When applied to soil in the laboratory, GA_3 also enhanced germination of dormant seed of numerous weeds (1, 2, 5, 11, 15, 23) including many of the same species whose germination was stimulated by AC-94377 (4, 8, 19), such as wild mustard. Dormant wild mustard seed were chosen as a model system to study the impact of AC-94377 on seed survival in the field because of this weed's economic importance in cereal and sunflower (*Helianthus annuus* L.) production in the northern Great Plains of the U.S. and Canada (20).

Few field studies demonstrate that germination stimulants actually speed the loss of dormant seed from the soil seedbank (reviewed in 8, 19). Most methods used to establish that germination stimulants decrease seed survival in the field are indirect. Weed emergence has been used most often for this purpose. Efficacy of germination stimulants to decrease survival of seed buried in nylon mesh packets has not been studied previously. Use of nylon mesh packets has the potential for improving control of variability and for screening candidate chemicals compared to sampling weed seed in soil from treated field plots. Seed packets also minimize seed predation by insects, birds, and mammals and allow a more complete accounting for buried seed than is possible when seed are mixed with soil and are later extracted from soil.

Seed dormancy in wild mustard in complex. Wild mustard embryos are fully formed at maturity and germination of newly mature seed increased when their seed coats were removed (11, 16), suggesting that the seed coat restricts germination in some way. Edwards (11) attributed loss of dormancy to decay of restrictive seed coats in the field, since seed stored dry in the laboratory remained highly dormant. Germination of seed recovered from soil decreased as burial depth increased (5 vs. 2.5 cm) (15). Progressively deeper burial of wild mustard seed in soil reduced germination of unearthed seed placed in petri dishes and imposed dormancy by inducing a light requirement for germination that could be overcome by exogenous application of GA_3 (15).

[1]Received for publication June 8, 1992, and in revised form December 19, 1992. Contribution of U. S. Dep. Agric., Agric. Res. Serv. Published with the approval of the Director, Agric. Exp. Stn., North Dakota State Univ. as J. Art. No. 2050.

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[3]Letters following this symbol are a WSSA-approved computer code from Composite List of Weeds, Revised 1989. Available from WSSA, 309 West Clark Street, Champaign, IL 61820.

Buried, undisturbed wild mustard seed were persistent in soil (14, 20) and had a half-life of 3 yr in England (12). In another study, 86% of undisturbed buried seed survived after 3 yr and 17% survived after 14 yr of burial (16), but shallowly buried seed were less persistent than more deeply buried seed. Fifty percent of buried wild mustard seed survived for 7 yr when left undisturbed under a smooth brome (*Bromus inermis* L.) pasture in Minnesota (28). However, only 3% of buried seed survived after 7 yr when the soil was tilled three times per year. In Canada, repeated tillage during a fallow-spring wheat-fallow rotation resulted in the loss of 94 to 99%, 0 to 1.5%, and 0.5 to 3.4% of buried seed in years one, two, and three, respectively, when seed production was prevented (6). Thus, periodic soil disturbance, as by tillage, enhanced the rate of loss of wild mustard from the soil seedbank (28), presumably by stimulating germination (3, 12).

The objectives of these experiments were to determine the impact of AC-94377 rate, application timing, and application frequency on wild mustard seed survival in the field; and determine whether nylon seed burial packets of different sizes interfered with the biological activity of AC-94377 on dormant wild mustard seed.

MATERIALS AND METHODS

AC-94377 effects on wild mustard seed survival in the field. The treatments were AC-94377 at 0, 1.7, and 3.4 kg ha^{-1} every fall for each of 4 yr, every spring, both every fall and spring, only the first fall, only the first spring, and only the first fall and spring after burial. A randomized complete block design with four blocks was used in a factorial arrangement and the experiment was repeated at one location (trials 1 and 2, respectively). Blocking was based on anticipated moisture gradients across the location due to snow accumulation. Trial 1 lasted from the fall of 1983 to 1987 and trial 2 lasted from the fall of 1984 to 1988. Pots were treated with AC-94377 on Sept. 14, 1983 (trial 1) and Sept. 14, 1984 (both trials), Sept. 11, 1985 (both trials), Sept. 16, 1986 (both trials), and Sept. 14, 1987 (trial 2). Pots were treated on Apr. 18, 1984 (trial 1) and 1985 (both trials), Apr. 21, 1986 (both trials), Apr. 17, 1987 (both trials), and Apr. 15, 1988 (trial 2).

Fully mature wild mustard seed from a presumably genetically diverse wild population were gathered near Fargo, ND, Aug. 4, 1983 for trial 1 and from the same location Aug. 3, 1984 for trial 2. Previous research conducted over several years demonstrated that freshly shed wild mustard seed are highly dormant and viable, as measured by high percent germination response to AC-94377 in petri dishes (8, 19). Air-dried wild mustard seed were stored in darkness at –15 C prior to packaging for burial in the field Sept. 13, 1983, for trial 1 and Sept. 21, 1984, for trial 2 to minimize afterripening and loss of initial dormancy. One hundred seed were packaged in nylon mesh[4] seed packets measuring 5 by 12 cm which were buried 1.4 cm deep in pots (7-cm diam by 7.5 cm tall) containing Fargo silty clay (fine, montmorillonitic, frigid Vertic Haplaquolls) with 2% sand, 47% silt, 51% clay, 3.9% organic matter, and a pH of 7.7. The coarse mesh packets were easily wetted and allowed free passage of water over the course of the experiment. Enough seed packets were buried so that four packets could be unearthed for each treatment in each of 4 yr. Pots containing seed packets were partially buried on the North Dakota State University Experimental Farm in Fargo. The trials were fenced and individual pots were covered with wide-mesh screening which prevented packet disturbance by rodents or birds but which allowed unrestricted infiltration of rainfall. Weeds were controlled by periodic application of glyphosate [*N* (phosphonomethyl)glycine] at less than 1.1 kg ha^{-1} around the pots.

AC-94377 synthesized by the method of Tanaka et al. (25) was applied uniformly to the soil surface with a pipette in 1 ml of acetone per pot at application rates of 1.7 and 3.4 kg ha^{-1}. The acetone evaporated within seconds of application. Exposure of wild mustard seed buried 2.5 cm deep in soil to acetone, acetaldehyde, and ethanol for 2 wk did not affect subsequent germination after removing the seed from soil (15).

Seed packets for both trials were retrieved from the field at yearly intervals on Sept. 14, 1984, Sept. 11, 1985, Sept. 16, 1986, Sept. 14, 1987, and Sept. 15, 1988 before treating some remaining seed with AC-93477 in the field. Seed were removed from the packets, washed free of soil, air dried, and stored in darkness at room temperature until germination was tested (usually within 7 d after unearthing). The number of intact, firm, hard seed surviving treatment was counted and the number of seed surviving over time (i.e., seed survival) was expressed as a percent of the initial number of seed buried (= 100), a widely accepted method in seed ecology and plant population biology. Since wild mustard seed do not have hard water-impermeable coats, they are likely to decay soon after death in soil, although this point remains to be studied.

Even though the main goal of the research was to determine the impact of seed treatment on seed survival, viability of surviving seed also was estimated by percent germination, although it was of only secondary research interest. The tetrazolium test for viability was found to be unreliable for determining viability of this small-seeded species.

Germination was expressed as a percent of the number of surviving seed unearthed each year. When seed were first buried, four groups of 100 seed each were germinated in 6-cm-wide plastic petri dishes containing two Whatman # 2 filter papers moistened with deionized glass-distilled water. Seed were germinated for 2 wk in an incubator in darkness at 20 C and 100% relative humidity. Wild mustard seed germinated best at 21 C when tested from 11 to 30 C (13, 14) and at 22 C when tested from 1 to 27 C (11).

Data were subjected to analysis of variance, and both linear and nonlinear least squares regression analysis were used to describe percent seed survival versus time for each

[4]Nitex monofilament nylon 480 micron mesh. Pesco, Inc., P.O. Box 24225, Minneapolis, MN 55424, U.S.A.

trial using Tablecurve version 3.0 software[5]. Data are presented by trial, frequency, and rate because there were significant two- and three-way interactions. The 20 nonlinear regression equations generated by Tablecurve with the greatest F-values were compared using the magnitude of both the F-value and R^2 (i.e., the proportion of variability due to the independent variable in the regression equation). Inspection of plots of residuals versus the independent variable was used to evaluate model adequacy. The slopes and intercepts of regression models are presented ± standard errors. All slopes and intercepts were significantly different from zero at $P < 0.01$, at least.

Effect of seed packets on wild mustard seed response to AC-94377. In this greenhouse pot experiment AC-94377 at 0 and 1.7 kg ha^{-1} was applied to seed packets on the soil surface and buried 1.25 cm deep. There were six seed packet treatments: no nylon[4] seed packets, small nylon seed packets (5 by 6.25 cm) either with or without soil, and large nylon seed packets (5 by 12 cm) either with or without soil. (See soil description above.) A completely randomized design with four replications was used with treatments in a factorial arrangement and the experiment was repeated.

Mature, dormant wild mustard seed were collected near Fargo, ND, Aug. 15, 1986, air dried, and stored at –15 C in darkness. Packets each containing 100 wild mustard seed were placed in rectangular plastic pots (12 cm wide by 16.5 cm long by 5.5 cm tall) (1-L vol) before spray treatment. AC-94377 (50% wp[6]) was applied to the soil surface with a hooded sprayer equipped with a flat-fan nozzle delivering 300 L ha^{-1} in one pass at 180 kPa and 1.7 kg ha^{-1} Feb. 26 and June 7, 1987 in repetition 1 and 2, respectively. Seed were top watered in the greenhouse after treatment to keep the soil surface continuously moist. Greenhouse temperatures averaged 24 and 15 C during the day and night, respectively. Natural daylight was supplemented with fluorescent lighting to provide a 14-h photoperiod. Fluorescent light intensity was about 25 W m^{-2} and photosynthetically active radiation of 125 μE m^{-2} s^{-1} at the pot surface measured at night. Four weeks after spray treatment, surviving seedlings were counted either on the soil surface or in the packets. Seedlings were unable to penetrate the seed packets. Then, remaining ungerminated seed were counted from packets or were sieved from the soil in packets (#20 sieves) and counted. Percent data were subjected to analysis of variance both directly and after arc sin square root transformation to minimize variance nonhomogeneity. Percent means ± standard errors are presented for the average of two repetitions of the experiment.

RESULTS AND DISCUSSION

AC-94377 effects on wild mustard seed survival in the field. AC-94377 at 3.4 kg ha^{-1} applied annually greatly reduced survival of shallowly buried, undisturbed wild mustard seed in the field compared to the untreated check (Figures 1 and 2). In both trials, AC-94377 applied in spring alone, fall alone, or both spring and fall reduced seed survival. However, in trial 1, both fall treatments were more effective than spring treatment alone, as indicated by progressively decreasing X-axis intercepts and increasing slopes for nonlinear regression equations for these respective treatments (Figure 1). Fall treatments in trial 1 could not be distinguished from one another. In trial 2 all three treatment timings reduced wild mustard seed survival equally well (Figure 2). No seed survived AC-94377 treatment applied annually in either spring alone or fall alone by year three in trial 2 (arrows in Figure 2), but some seed survived these treatments in trial 1 (Figure 1). No seed survived annual AC-94377 treatment applied in both fall and spring beyond year three in either trial, unlike the untreated seed.

For the sake of brevity, only data for AC-94377 applied in fall alone are presented in the discussion of the impact of phthalimide rate on wild mustard seed survival (Figure 3). The impact of AC-94377 applied in fall alone on wild mustard seed survival was intermediate between treatments applied in spring alone or both fall and spring. As expected, annual AC-94377 application for 4 yr reduced wild mustard seed survival more than treatment in only the first year (Figure 3). In both trials, AC-94377 applied each fall for 4 yr greatly reduced wild mustard seed survival. When AC-94377 at either rate was applied only the first fall after seed burial, it failed to reduce seed survival in trial 1 but reduced seed survival in a rate-dependent fashion in trial 2 (Figure 3). (Data for AC-94377 at 1.7 kg ha^{-1} not presented.)

Survival of AC-94377-treated and untreated wild mustard seed was a negative exponential (log-linear) function of time in trial 1 (Figure 1), as described elsewhere (7, 21). In trial 2, the seed survival curve of the untreated check was a negative arithmetic (linear) function of time (Figure 2) but became a negative exponential function of time following AC-94377 treatment. Results of several seed survival studies have been published on wild mustard, yet only Edwards (11) suggested that this species' seed survival was a negative exponential function of time, although Warnes and Andersen (28) presented data which could probably have been well described using this relationship.

Differences in seed survival of the untreated check between years. The greatest absolute difference in seed survival for the untreated check between trials 1 and 2 occurred by the first fall 1 yr after burial (Figures 1 and 2). The untreated check in trial 1 experienced a larger decrease in seed survival by year one than in trial 2. Seed survival of the shallowly buried untreated check was less than most reports (12, 14, 28) but similar to seed survival after periodic tillage (6).

There are several explanations for observed differences in shapes of seed survival curves for the untreated check between trials (Figures 1 and 2). Differences in seed survival for the untreated check between trials could be due to: genetic differences between seed lots used for each trial; differences in initial dormancy between seed lots due to differences in

[5]Jandel Scientific, 65 Koch Rd., Corte Madera, CA 94925, U.S.A.

[6]Formulated AC-94377 provided by Dr. Pavlistra, American Cyanamid Co., Agric. Res. Div., P. O. Box 400, Princeton, NJ 08540, U.S.A.

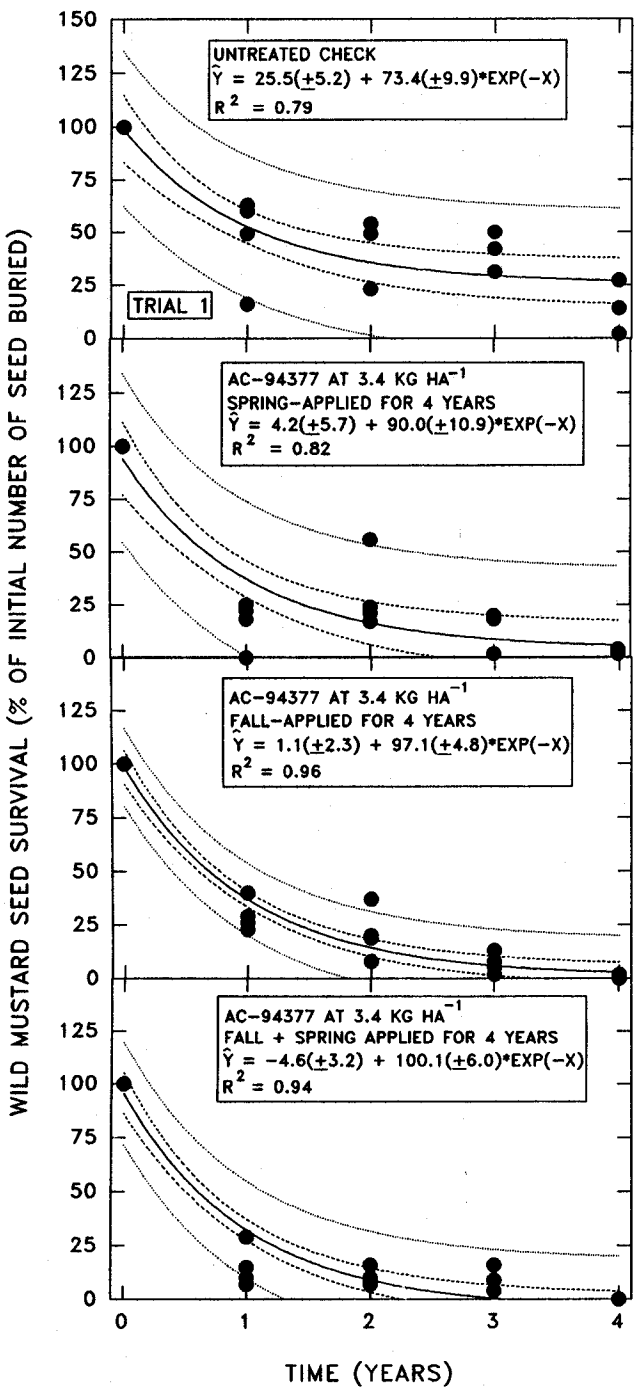

Figure 1. The effect of timing and frequency of application of AC-94377 at 3.4 kg ha^{-1} on wild mustard seed survival over time in trial 1. AC-94377 was applied either in spring or fall alone or in both spring and fall for each of 4 yr. Coefficients for equations with ± standard errors in parentheses and data points (circles) are presented for equations, as well as 95% confidence intervals (dashed lines).

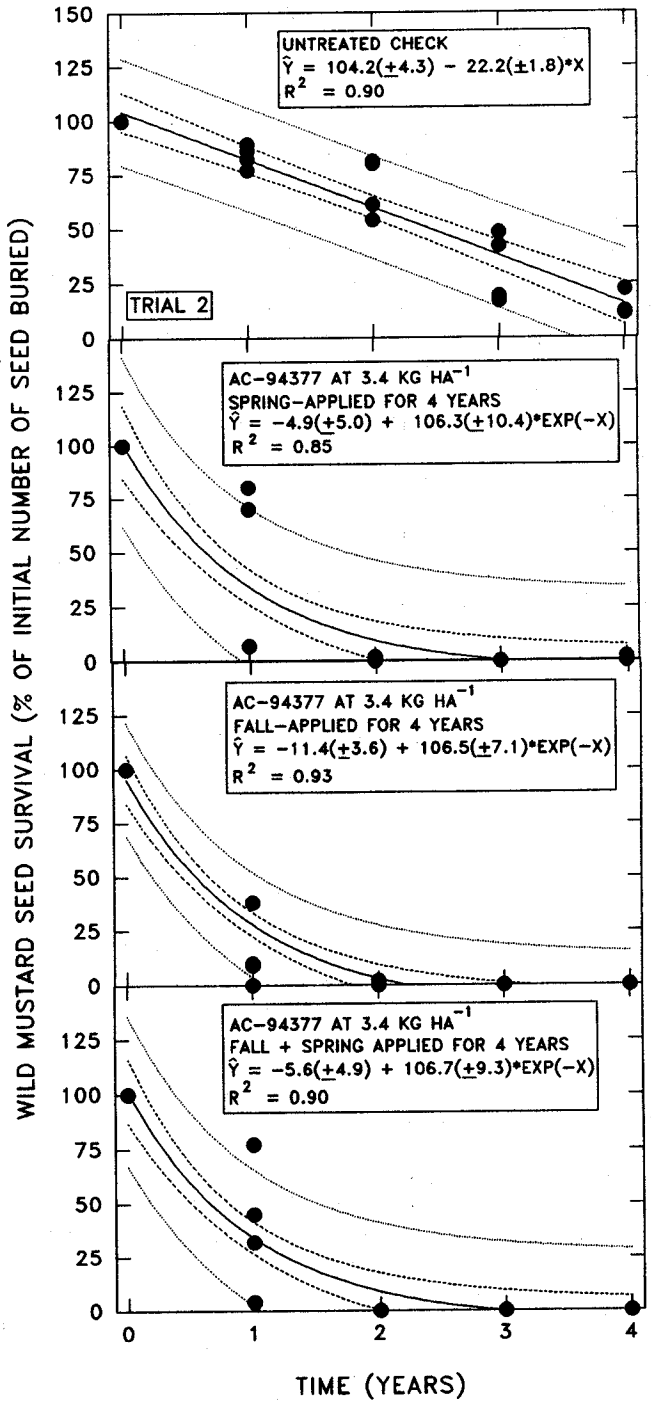

Figure 2. The effect of timing and frequency of application of AC-94377 at 3.4 kg ha^{-1} on wild mustard seed survival over time in trial 2. AC-94377 was applied either in spring or fall alone or in both spring and fall for each of 4 yr. Coefficients for equations with ± standard errors in parentheses and data points (circles) are presented for equations, as well as 95% confidence intervals (dashed lines).

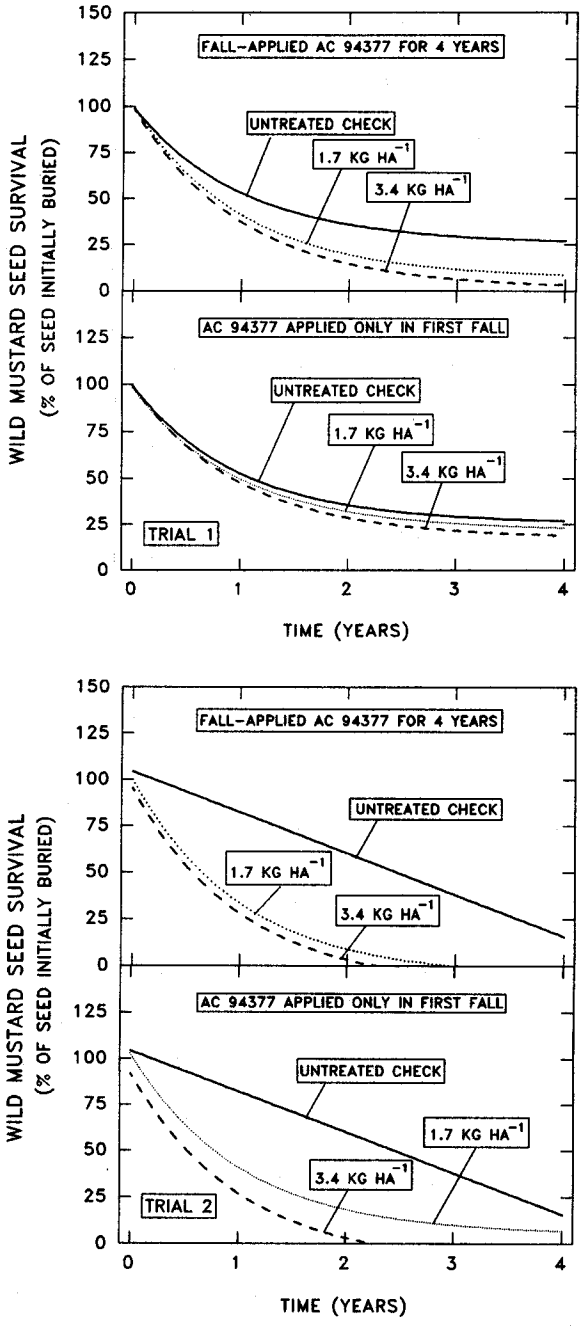

Figure 3. The effect of frequency of application of AC-94377 at 3.4 kg ha^{-1} in fall (first fall only or each of 4 falls) on wild mustard seed survival over time. In trial 1, the equation for the untreated check was Y = 25.5 (± 5.2) + 73.4 (± 9.9)·EXP (–X) (R^2 = 0.79), where Y = wild mustard seed remaining as a percent of the initial number buried and X = time in years. Coefficients for equations with ± standard errors in parentheses are presented. For AC-94377 at 3.4 kg ha^{-1} applied in each of 4 falls, the equation was Y = 1.1 (± 2.3) + 97.1 (± 4.8)·EXP(–X) (R^2 = 0.96), and for treatments applied only in the first fall, the equation was Y = 17.4 (± 3.6) + 81.9 (± 7.4)·EXP(–X) (R^2 = 0.87). In trial 2 the equation for the untreated check was Y = 104.2 (± 4.3) – 22.2 (± 1.8)·X (R^2 = 0.90). For AC-94377 at 3.4 kg ha^{-1} applied in each of 4 falls, the equation was Y = –11.4 (± 3.6) + 106.6 (± 7.1)·EXP(–X) (R^2 = 0.93) and for treatments applied only in the first fall, the equation was Y = –11.0 (± 4.4) + 102.8 (± 8.8)·EXP(–X) (R^2 = 0.88).

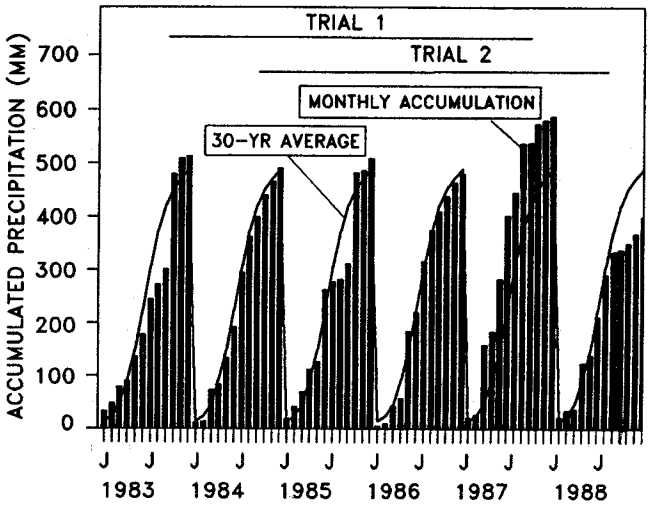

Figure 4. Monthly accumulated precipitation (= solid vertical bars) for trials 1 and 2 and the 30-yr average accumulated precipitation (= cubic spline line). Weather data were gathered at Hector International Airport approximately 1 km north of the experimental site. The period over which trials 1 and 2 were conducted is indicated by horizontal lines.

environmental conditions during seed maturation between trials; or differences in germination, emergence, and establishment over time because environmental conditions varied between trials during peak periods of germination.

Rainfall was similar to the 30-yr average in the first spring following fall burial in both trials (Figure 4). Rainfall in spring did not limit wild mustard emergence and thus was not responsible for enhanced seed survival in trial 2, because spring emergence [31.6 ± 2.9% (mean ± standard error)] in 1985 (trial 2) was somewhat greater than spring emergence [24.0 ± 4.2% (mean ± standard error)] in 1984 (trial 1) during the first spring following fall burial. Wild mustard is a summer annual in most temperate climates (20). Most emergence occurs during March and April but may continue into summer under favorable environmental conditions (12, 22). Emergence in fall immediately after burial was very limited [1.9 ± 0.5% and 2.9 ± 0.4% (mean ± standard error)] in trials 1 and 2, respectively. Freshly shed seed of wild mustard germinated poorly in the first fall (1%) after dispersal compared to the subsequent spring (≤ 75%) (14). Seed germination is thought to be the major cause for seed loss from the buried seedbank (7, 21). Seed packets prevented seed predation by rodents, birds, or insects (18, 27) in this experiment, but microbial attack was not prevented.

Seed used for both trials were gathered from wild populations at the same location in two successive years, making it unlikely that genetically different weed populations were sampled. However, seed used for trial 1 matured during a period of summer drought, whereas those used for trial 2 matured under more normal rainfall conditions (Figure 4). Percent germination of untreated wild mustard seed was 11.5 ± 2.2% and 1.0 ± 2.2% (mean ± standard error) at the time of

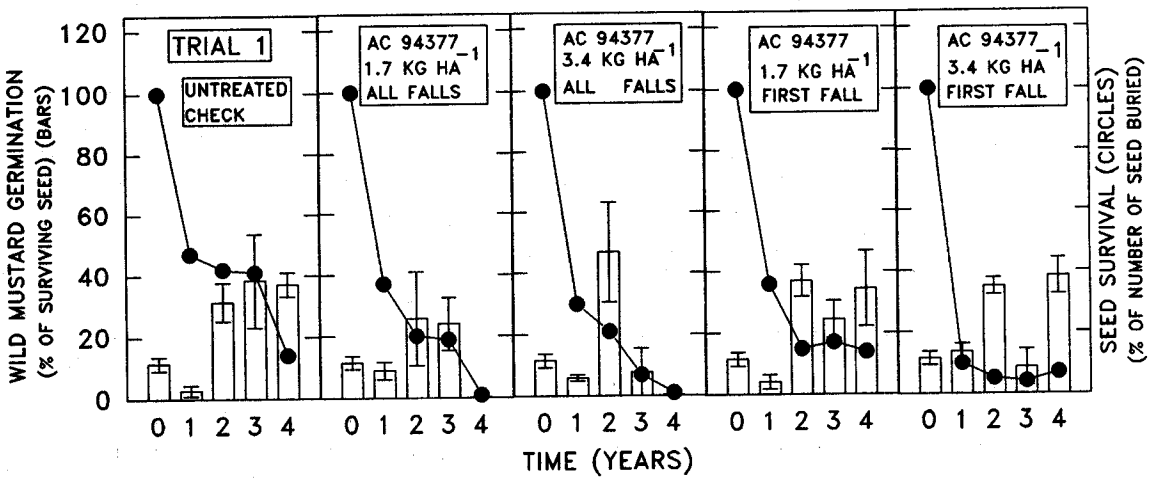

Figure 5. The effect of AC-94377 rate (0, 1.7, and 3.4 kg ha^{-1}) and frequency of application in fall (first fall only or each of 4 falls) on percent germination of surviving wild mustard seed in fall over time in trial 1. Survival (solid circles) is expressed as a percent of the initial number of seed buried whereas germination (bars) is expressed as a percent of seed surviving in each year (thus, sample size for percent germination decreases as fewer seed survive over time). Means ± standard errors are presented.

burial in trials 1 and 2, respectively, but slightly more seed emerged during the fall and spring following burial in trial 2 than in trial 1 (reported in previous paragraph). Differences in summer rainfall during seed maturation leading to differences in phenotypic differences likely account for the differences in seed survival observed between trials (Figures 1 and 2).

Seed survival over time for a species is best described by families of curves, rather than one class of functional relationships (i.e., negative exponential) (17). The shape of seed survival curves of untreated checks (Figures 1 and 2) is probably not a species characteristic and may reflect the interaction of a wild mustard's seed phenotype and environment during seed maturation and after dispersal. Infrequent sampling (i.e., once per year in this seed survival study) combined with sampling error may mask more complex seed survival curves, as observed for jointed goatgrass (*Aegilops cylindrica* L.) seed survival when seed were sampled at biweekly intervals over 2 yr (9) compared to less frequent yearly sampling (10).

Few seed survival studies are repeated over time. Londsdale (17) predicted that year-to-year differences in shapes of seed survival curves may be due to differences in the frequency distribution of different seed phenotypes. However, fluctuations in year-to-year environment during seed maturation (Figure 4) may also contribute to differences in the shapes of wild mustard seed survival curves for different lots of seed buried in different years (Figures 1 and 2).

AC-94377 effects on percent germination of surviving wild mustard seed. After the number of wild mustard seed surviving each fall was counted, their germination ability expressed as a percent of surviving seed (not the initial number of seed buried) was determined under reproducible, favorable environmental conditions in a dark incubator. In trial 1, untreated seed surviving into year one germinated less than at burial, but percent germination of surviving seed increased after year one (Figure 5). In trial 2, untreated seed germination increased progressively the longer seed were buried (Figure 6). Percent germination of remaining untreated seed was greater in trial 2 than trial 1 in 3 of 5 yr (years one, three, and five, but not at the start).

Wild mustard germination reportedly increased the longer seed were buried (11, 16), with germination of shallowly buried, undisturbed seed increasing from 3 to 80% over 3 yr (16). In a second study, seed buried 1.3 cm deep germinated 20, 50, and 80% after 8, 18, and 28 mo of undisturbed burial, respectively (11). Edwards (11) attributed loss of dormancy to decay of restrictive seed coats in the field, since seed stored dry in the laboratory remained highly dormant.

In both trials, percent germination of seed treated with AC-94377 at 3.4 kg ha^{-1} surviving into year one was greater than untreated check seed for year one (Figures 5 and 6). Percent germination of seed surviving fall AC-94377 treatments also was much greater in trial 2 than trial 1 in year one for AC-94377 at both rates and in year two for AC-94377 at 1.7 kg ha^{-1}.

Effect of seed packets on wild mustard seed response to AC-94377. Wild mustard seed buried 1.25 cm deep did not respond to AC-94377 as much as surface-lying seed in the greenhouse, as measured by seed survival 1 mo after treatment. Emergence of buried seed was limited, leading to greater than 70% seed survival for all treatments irrespective of seed packet (Figure 7). Daily watering was expected to transport AC-94377 to the shallowly buried seed (8), but 1 mo may not have been sufficient time to transport AC-94377 to the burial depth of the seed to influence dormancy.

AC-94377 speeded the loss of surface seed without nylon seed packets more than seed buried in packets without soil 4

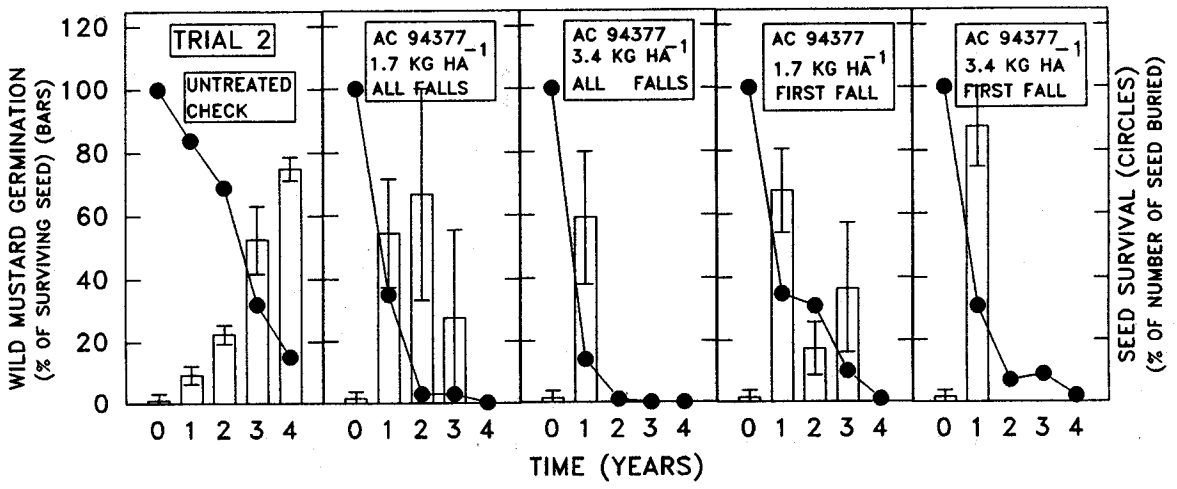

Figure 6. The effect of AC-94377 rate (0, 1.7, or 3.4 kg ha^{-1}) and frequency of application in fall (first fall only or each of 4 falls) on germination of surviving wild mustard seed in fall over time in trial 2. Survival (solid circles) is expressed as a percent of the initial number of seed buried whereas germination (bars) is expressed as a percent of seed surviving in each year. (Thus, sample size for percent germination decreases as fewer seed survive over time.) Means ± standard errors are presented.

wk after treatment (Figure 7). When either sized seed packets contained soil, fewer seed survived than without seed packets or from packets without soil. More surface wild mustard seed established in response to AC-94377 at 3.4 kg ha^{-1} from large than small seed packets either containing or not containing soil (Figure 8).

Apparently, the nylon packet environment was unfavorable for wild mustard seedling establishment but allowed emergence (Figure 8). More untreated surface seed established (≥ 25%) without seed packets than in seed packets 4 wk after the start, even when soil was added to packets (Figure 8). Adding soil to either large or small seed packets on the soil surface did not greatly increase establishment of untreated seed above that observed from seed packets of comparable size without soil. Because the sum of the surviving seed (Figure 7) and established seedlings (Figure 8) did not account for the total number of seed buried, many seed presumably germinated and died. Empty seedcoat fragments and decaying remains of germinated seedlings were observed. This was more noticeable for the small than large seed packets filled with soil.

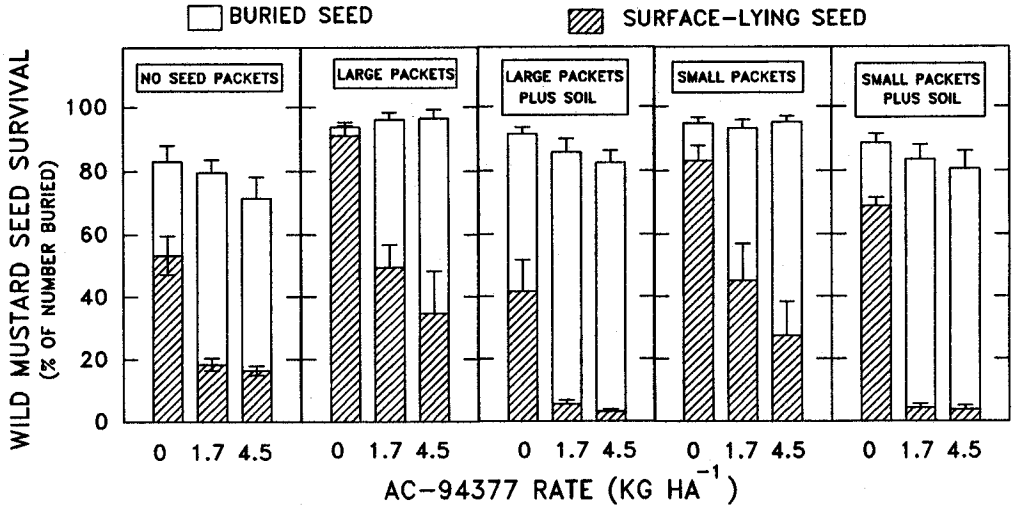

Figure 7. The effect of AC-94377, depth of burial, and nylon seed packets on wild mustard survival 4 wk after treatment. Surface-lying seed are represented by hatched bars and seed buried 1.25 cm deep are represented by open bars. Means ± standard errors are presented for the average of two repetitions of the experiment.

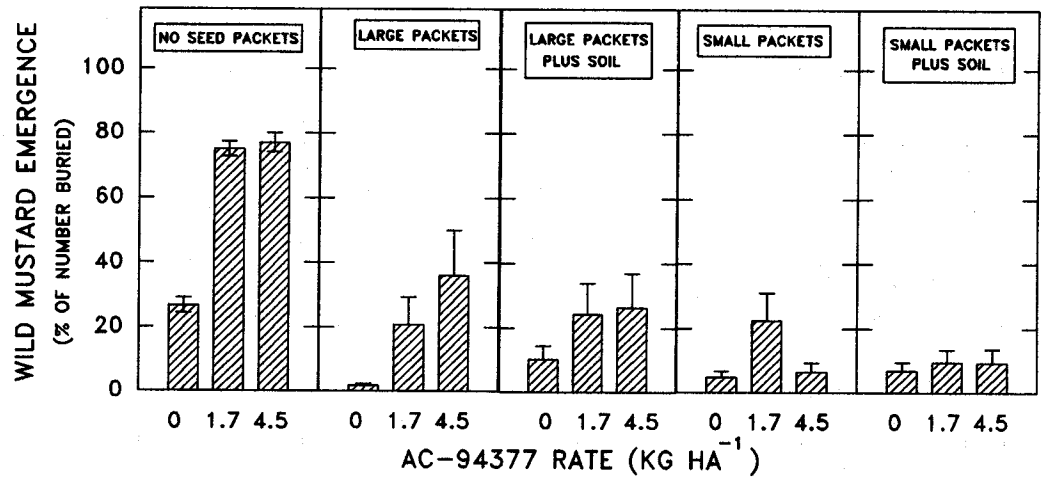

Figure 8. The effect of AC-94377 and nylon seed packets on wild mustard germination and establishment from the soil surface. No seed established in buried seed packets (data not presented). Means ± standard errors are presented for the average of two repetitions of the experiment.

This field seed persistence study is unique in that it was repeated in time and examined the impact of repeated chemical treatment on seed survival over time. The kinetics of seed survival for untreated seed varied between trials and deserves further long-term study. This research demonstrates that AC-94377 can speed the loss of shallowly buried, undisturbed weed seed in the field. Thus, it is most likely that AC-94377 would have the greatest potential for decreasing weed seed levels in reduced-tillage systems in which most seed are buried close to the soil surface. Studies of seed survival in nylon seed packets have potential for "screening" germination stimulants under field conditions over a relatively short burial period of 3 to 4 yr. Greenhouse experiments demonstrated that large nylon seed packets should be used in burial studies, as was done in this research, and that soil should be included with the seed in the burial packets. Nylon seed packets without soil may limit seedling establishment. Small nylon packet size may also limit seed access of individual seeds to phthalimide; when seed density is high in small seed packets, phthalimide uptake by individual seed may become restricted. The potential biological activity of germination stimulants, such as AC-94377, to reduce the seedbank may be demonstrated better by measuring seed survival under field conditions rather than following seedling emergence over time. Counting emerged seedlings seriously underestimates the effectiveness of germination stimulants to deplete the soil seedbank because emerged seedlings are only a small proportion of the total seedbank (21). It was not the intent of this research to prove the commercial feasibility of this approach for ridding fields of weed seed.

ACKNOWLEDGMENTS

We thank Roland Hoerauf and Ronald Wien for their technical assistance.

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